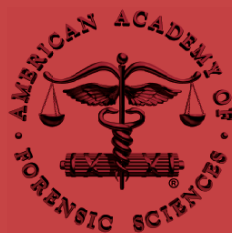


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*American
Academy
of Forensic
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*66th Annual Scientific Meeting
Seattle, WA
February 17-22, 2014*



AMERICAN ACADEMY OF FORENSIC SCIENCES

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PROCEEDINGS

of the American Academy of Forensic Sciences
66th Annual Scientific Meeting

The *Proceedings* of the American Academy of Forensic Sciences is an official publication of the American Academy of Forensic Sciences (AAFS). It is devoted to the publication of the abstracts of technical oral papers and posters presented at the AAFS annual scientific meeting. These include various branches of the forensic sciences such as pathology, toxicology, physical anthropology, psychiatry, immunology, odontology, jurisprudence, criminalistics, questioned documents, digital evidence, and engineering. Similar submissions dealing with forensic oriented aspects of the social sciences are also included.

Please note that some of the abstracts included in the *Proceedings* deal with topics, results, and/or conclusions which are controversial. The publication of abstracts does not imply that the AAFS, its sections, or the individual section program chairs/committee members have verified or agree with the studies, results, and/or conclusions of each abstract. During the process of planning a scientific program, it is impossible to "peer-review" each abstract and presentation to the degree that is accomplished during manuscript review. Abstracts and presentations are accepted, in part, so that they can be critiqued and reviewed by other scientists. Thus, a forum is created to discuss controversial issues.

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Proceedings

of the American Academy of Forensic Sciences

February 2014

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SPECIAL SESSIONS



SEATTLE 2014

SEATTLE 2014

S1 Fostering the Next Generation of Forensic Scientists: Mentoring in the 21st-Century

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After attending this presentation, attendees will have a better understanding of mentorships, training, young scientist professional committees and other educational development opportunities available within the various disciplines of the eleven sections of the American Academy of Forensic Sciences (AAFS).

This Interdisciplinary Symposium will impact the Forensic Science community by informing attendees of opportunities that will enhance their professional development and encourage networking and mentoring of new members of the field.

Description: Tomorrow's forensic science leaders are in our classrooms and laboratories today. Education of the next generation of forensic scientists, the continuing professional development and training of current practitioners, and those in the legal community is our collective responsibility and of paramount importance to the future of our profession. In the spirit of the AAFS 66th Annual Scientific Meeting theme, Forensic

Science Education and Mentorship: Our Path Forward, this symposium brings together each section of the Academy to discuss how mentorship, education, and partnership can advance the field of forensic science for the future.

Criminalistics: The 2,833 members of the AAFS Criminalistics Section represent a wide variety of forensic science

disciplines and a wealth of hands-on forensic science knowledge, skills, and abilities. The members are actively engaged in research, education, training, continuing education, mentoring, and hands-on projects and programs. The Forensic Science Program at San Jose State University (SJSU) is an example from the Criminalistics Section of AAFS that has developed educational opportunities and research mentorship programs. First, there are two formal undergraduate program offerings in Forensic Science: Bachelor of Science in Forensic Science (BSFS) with a Concentration in Biology or a BSFS with a Concentration in Chemistry. Both programs were developed to meet FEPAC educational standards where students gain knowledge through lectures and reading, hands-on activities, and laboratories and develop critical thinking through scientific experimentation. Second, has established partnerships with local, regional, national, and international crime laboratories and agencies that have provided mentoring and internships to students. Students who conduct research internships are mentored by experts in their respective fields and some conduct their research at crime laboratories and agencies offsite. The AAFS supports mentoring through the YFSF Mentor Program available for participation by all section members. The Mentor Program provides opportunities to mentor new forensic scientists. Third, SJSU is actively involved in middle- and high-school science and math education through developing and delivering Crime Scene Investigation (CSI) Summer Camps and AAFS Forensic Science Educational Conferences. The speakers and staff at these camps and conferences are drawn from the AAFS Criminalistics membership and other experienced forensic scientists and practitioners providing hands-on activities that the teachers can take back to their own classrooms.

Digital & Multimedia Sciences: Not only is the Digital & Multimedia Sciences Section the newest section in the Academy, but the field of digital forensics is one of the newest in the constellation of the forensic sciences. It has its roots in the investigative need to recover, extract, and analyze information stored and transmitted in binary form. This presentation will describe how the AAFS has, and continues to be, an important catalyst in the continuing development of the sciences, as well as a forum for developing the leaders of the next generation of digital forensics practitioners. The presentation will focus on the role of the Academy in the discipline's development of standards, educational requirements, and the Kuhnian notion of a scientific discipline. The speaker will describe how a relatively small group of pioneers, within scarcely a generation, laid a foundation for a new scientific discipline. The presentation will describe the current state of the discipline and how three "generations" of digital forensic practitioners are cooperating, collaborating, and mentoring each other even as the technical challenges and volume of data continue to mushroom. Standards bodies, professional certifications, and accreditation of both laboratories and educational institutions will be discussed.

Engineering: Engineering is a broad spectrum discipline with a number of renowned specialties. But the fundamental link is the foundation of scientific discipline that graces the engineering profession, no matter what the specific discipline. Forensic engineering is the particular ability to apply one's professional skills to the matters of legal inquiry. The mentor in forensic engineering looks to first evaluate the mentored engineering candidate for potential of exemplary engineering practice ability and professional

ethics, and, secondly, to ensure there is a clear understanding and ability to investigate engineering issues as related to the law. The mentor candidate is then exposed to engineering problems involving actual forensic cases. The examination of case histories is repeated until the candidate is well-versed in independent analysis of the issues and in the ability to clearly explain engineering principles and solutions to a wide variety of forensic problems in both written and verbal forms. A good candidate in forensic engineering is one who is confident in his/her engineering abilities and can comfortably carry the essence of a teacher of the engineering principles to the legal forum, without arrogance. The candidate is encouraged to become involved with the AAFS, as well as other professional organizations that build the person's currency in the specifics of the candidate's specialty, and encourage awareness of the particulars of the legal aspects. The mentoring may continue through philosophical discussions after the engineer provides deposition and court testimony independent of the mentor. The mentor's role is to encourage, support, and provide critique as a continuing building of the profession.

General: The General Section of AAFS is one of the most diverse sections of the Academy with regard to the spectrum of membership disciplines. All disciplines offer informal open dialogue in an effort to increase education, qualifications, and experience for forensic scientists. For those disciplines that do not offer formal mentorship programs, practitioners are available to provide individual oversight to ensure that critical skills are taught and utilized appropriately. Outreach programs begin at the grade school level. An example is the demonstration of various forensic techniques at Girl Scout Camps by AFOSI agents to assist girls in earning their investigator badge. The girls gain knowledge about processing crime scenes and learn the requirements for becoming law enforcement agents, hopefully sparking their interest for future careers. Another avenue of outreach is through the use of technology to increase forensic science exposure. Technology allows for the convenience of opportunities. One section member has developed a popular weekly webcast TV show titled ForensicWeek.com. The talk show highlights different forensic topics presented by international forensic scientists and investigators. The live chats with practitioners provide synergy between experience, accurate scientific methods, and modern technology.

Jurisprudence: Five years after the NAS Report, forensic science is finally starting to make significant headway in the legal community. From introductory classes that gently introduce law students to the concept of questioning the strength and meaning of forensic sciences, such as courses called "Wrongful Convictions," to joint degree programs which allow a student to gain both a JD and an MS in forensic science, the face of forensic education in law schools is changing. Likewise, seminars on forensic science issues directed at the legal community are becoming more prevalent and more popular. As forensic divisions of public defenders' offices expand and settle in, opportunities for law students to clerk in a forensic setting before taking the bar exam puts them in a unique position post-passage of the bar exam. Finally, it is no longer unheard of (or forbidden) for a public defender to sign up for and attend a forensics class geared toward law enforcement. These are some of the many growth opportunities for lawyers interested in competently handling forensic science evidence.

Odontology: The American Board of Forensic Odontology offers workshops in conjunction with the Academy meeting each year. They are focused in these areas: dental age estimation, bitemarks, human identifications, mass fatality incidents, human abuse, and civil litigation. These workshops provide education, instruction, and hands-on casework in a smaller setting conducive to individualized feedback and mentoring. When successfully challenged, the workshops can provide casework that can be applied to the credentials necessary to challenge the American Board of Forensic Odontology.

Pathology/Biology: Mentoring in forensic pathology is a long-term endeavor. The community of forensic pathologists is so small and the need so great. The recruitment, development, and retention of candidates is critical to the future of death investigation in the United States. Early outreach is the first step for forensic pathologists seeking opportunities to access students in the biomedical sciences, as well as medical students, to expose them to the world of medicine and forensic pathology, and to promote relationships with the students early in their careers. The next step is to entice medical students into elective rotations so that they can observe and participate in death investigations and autopsy pathology with hopes that the experience will lead to residencies in pathology. By modeling the professionalism, ethics, spirit of scholarly inquiry and research, and the continuing education required of a career in forensic pathology, the forensic pathologist demonstrates how satisfying the in-depth study of death and its circumstances can be as a lifetime endeavor. Mentoring requires special attention to communicating the philosophy of practicing medicine on the dead — how daily exposure to death and violence inspires the melding of medicine and justice, medicine and the saving of lives of populations through the public health model and how it provides compassionate service to survivors. Even after training is completed, senior pathologist mentors consult with their trainees on cases and joint projects, and they cooperate in activities to promote the benefits of competent death investigation and forensic pathology. In such a small community of specialist physicians, relationships are strong and durable over the years.

Physical Anthropology: The research and education priorities in our major research institutions are being reconceptualized by the principles of "engaged scholarship," "in-service learning," "global citizenship," and "sponsored research" — concepts which fit well within forensic anthropology. Our pedagogical approach includes hands-on service learning and community engagement where students learn methods by doing actual casework; work with professional law enforcement through continuing education and professional development; and, design research questions based on problems arising during casework. For post-graduates and post-doctorates in forensic anthropology, there are a number of professional development and training programs available through medical examiner offices such as the Houston Medical Examiner's Office, Pima County OCME in Arizona, and the Visiting Scientist Program offered by the New York City OCME. Other academic programs have also been partnered with medical examiner offices such as the ICFahr consortium at the University of South Florida and the military training such as the JPAC Forensic Science Academy.

Psychiatry & Behavioral Science: Psychiatry & Behavioral Science Section members are currently involved in numerous types of mentorship, both generally and through involvement in specific AAFS activities and other activities. From a "general" mentorship standpoint, more senior section members solicit the trainees (particularly post-doctoral students, medical students, residents, and fellows) and early career mental health professionals from their respective academic institutions to join and become actively involved with AAFS. New members are encouraged to identify a mentor who can assist them in achieving their career goals and, if they are interested, gaining visibility and advancing in the section. From a "specific activity" mentorship perspective, section members are involved in numerous activities in both AAFS and outside organizations. Members of the section collaborate with outside national, regional, and local professional and volunteer organizations such as the American Academy of Psychiatry and Law's (AAPL's) Liaison with Forensic Sciences Committee and AAPL's Child and Adolescent Psychiatry Committee and the South Central Scholars Program.

Questioned Documents: Within the field of Forensic Documents (a.k.a. Questioned Documents) there has been a

noticeable increase in recent years in grant-funded research projects aimed at strengthening the scientific underpinnings of this discipline. Presentations and workshops at national and international conferences have seen a shift away from anecdotal case studies toward a platform focused more on research-oriented projects. Collaborations between the academic community and Forensic Document Examiners (FDEs) are also expanding, thereby generating additional opportunities for mentorships, internships, research, and continuing professional development. While some organizations, groups, and agencies have established programs to address education, mentorships, internships, research, continuing education/professional development, and certification relating to FDEs, many lack the resources to seriously maintain these worthy goals. Regardless of financial constraints, there are numerous ways to participate in these activities by utilizing the network of dedicated professionals which currently fill the ranks of the profession. The advancement of this discipline will continue to rely upon the tireless efforts of those individuals within the profession to work not only with other forensic scientists but also with non-forensic scientists, to share information and provide opportunities for others so that the next generations is properly equipped to continue to "raise the bar."

Toxicology: The Toxicology Section has a long history of mentorship of new toxicologists and opportunities for training, research, fellowships, continuing education, and awards and recognition of young talent. The section sponsors workshops at each annual meeting that provide not only educational knowledge, but fosters interaction between toxicology fellows, members, and student applicants. The Toxicology Section also has a highly active joint Drugs and Driving committee with the Society of Forensic Toxicologists (SOFT) that is always open and welcomes new members. This committee meets at each annual meeting and provides many resources, such as guidelines to the most important literature for testimony. The section has the June K. Jones Award that provides funding for research projects by new toxicologists and enables them to attend a meeting and present their findings. In addition, the Irving Sunshine Award is prestigious recognition of a young toxicologist's research and publications early in his/her career. Both of these awards introduce outstanding young toxicologists to the membership at a special evening reception in their honor and provide opportunities to further their careers. During the meeting, the Open Forum encourages young toxicologists to ask questions about issues, problems, procedures, new drugs, etc. in an informal and fun setting. These interactions frequently lead to summer internships, doctoral training opportunities, and potential job recruitments. The Toxicology Section has benefited from a large international membership that furthers our breadth of experience and informs us of new drug trends like the influx of new designer drugs that frequently are first encountered in Europe. These interactions are highly valuable and have led to visiting scientist programs in U.S. and international laboratories. The Toxicology Section membership overlaps extensively with SOFT and the International Association of Forensic Toxicologists (TIAFT), California Association of Toxicologists, and other professional groups. During the presentation, all of the resources of these sister organizations will be highlighted and how young toxicologists can take advantage of the many educational courses, fellowships, and experiences available to them.

Mentorships, Networking, Professional Development

S2 The World of Forensic Science: The Future of Education and Research

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After attending this session, attendees will have a better understanding of future trends in forensic science. Speakers will discuss unique casework as well as educational and career opportunities available. In addition, attendees will learn about casework and research that has been done by their peers at the Bring Your Own Slides and Poster sessions. The skills of including networking, personal branding, certification, and career preparation will also be discussed.

This session will impact the forensic science community by demonstrating the future trends in forensic science through casework, education, and mentorship opportunities. In addition, this session will provide young professionals with the tools needed to successfully contribute to the forensic science field.

For more than a decade, the Young Forensic Scientist Forum (YFSF) has provided a program for a group of students in both undergraduate and graduate programs, and forensic scientists with less than five years of professional experience. The goal of YFSF and the special session is to provide the participants the tools required to be a success in the field of forensic science. The session allows participants to interact with their peers as well as the professional speakers and to build professional relationships that can span a career.

Special session topics provide attendees with a broad outlook at the many opportunities in the field of forensic science. In addition to the special session, YFSF offers two opportunities for young forensic scientists to present their own work or research, the first being the YFSF Bring Your Own Slides (BYOS) session and the second, the YFSF Bring Your Own Posters (BYOP) session. In addition, the Forensic Sciences Foundation's Emerging Forensic Scientist Award Winner is always invited to present his/her award winning paper.

For the AAFS 66th Annual Scientific Meeting in Seattle, WA,

the YFSF Special Session will present: *The World of Forensic Science: The Future of Education and Research*. The special session will be held on Tuesday, February 18, 2014, and will include speakers who will discuss the various applications of education and research and the vast career paths within the field of forensic science by presenting their unique casework experiences and career choices. Through the presentations, speakers will demonstrate the different paths a forensic student or scientist can choose to take. The session will include speakers from many of the AAFS sections, highlighting the many opportunities the field may offer. Lunch is provided to both attendees and speakers who are registered for the special session.

Following the Tuesday session, the YFSF BYOP Session will be presented in the evening, giving young professionals the opportunity to showcase current cases and research in a poster format.

The annual YFSF BYOS Session takes place the evening of Wednesday, February 19, 2014, and will include presentations from students and new forensic scientists. YFSF does not require presenters of YFSF BYOS and BYOP to be members of AAFS and does not require they attend the special session but it is encouraged that they do so. The program will conclude Thursday, February 20, 2014, with the annual YFSF Breakfast Session which includes a resume review panel.

Navigating Success: Career Skills for the Journey — maintaining the focus on developing professional skills for the next generation of forensic professionals, the YFSF Breakfast Session will include established scientists presenting information to young forensic scientists and assisting them in creating successful resumes. Representatives from education, professional organizations, and various career stages will present on career skills — including networking, personal branding, certification, and career preparation — that assist emerging scientists as they journey along the forensic path. After the presentations, attendees will have the opportunity to interact with experienced AAFS members, receiving resume assistance and feedback.

YFSF, Education, Research



BREAKFAST SEMINARS



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BS1 Anatomy of a Conspiracy Theory — Death and Life, Myths and Realities

John U. Downs, BA, Charleston Auto Glass, 7 Amy Eley Drive, Charleston, SC 29407; and J.C. Upshaw Downs, MD*, GBI ME, 925 A Mohawk Drive, Savannah, GA 31419

After attending this presentation, attendees will understand the nature of various conspiracy theories and how they come to exist by using an overview and targeted case-study format to examine how evidence comes to be interpreted and the importance of context.

This presentation will impact the forensic science community by providing a knowledge base of the nature of conspiracy theories as a whole by a broad overview of the subject and a case study of the clues surrounding the alleged death of a famous public figure in hopes of better understanding how such theories are formed and thereby learning to better handle such events in casework.

The proliferation of conspiracy theories has been attributed to the interaction of enlightenment, polarization, and explanation. A commonality is an understandable theory appealing to “the enlightened” that the uninformed cannot or do not acknowledge. The mantra in detecting such plots is cui bono (who benefits?). Pseudoscience, terrorism, public figures, and governmental involvement are frequent springboards. Recent examples include governmental 9/11 collusion, 2004 tsunami, Presidential-associate executions, Princess Diana’s death, “Black Ops” helicopters, manufactured HIV, etc. Popular literature has reached back over two millennia to challenge the nature of the relationship between Jesus and Mary. Alleged cover-ups include the Pearl Harbor attack, Holocaust denial, Hitler’s faked death, Roswell’s Area 51, and innumerable other UFO legends. Famous conspiracies of the 1960s range from the “faked” moon landing to the Vietnam War to the John Birch Society-Communism controversy.

Perhaps the most familiar “plot” involves the assassination of President John F. Kennedy in 1963. Alleged political murders of the period include Robert F. Kennedy, Martin Luther King, Jr., and Malcolm X. Other famous popular figures of that era are not immune — the deaths of Marilyn Monroe, Sam Cooke, Brian Jones, Jimi Hendrix, Janis Joplin, and Jim Morrison have reported unusual elements, but all have in common that they actually occurred. In contrast, Elvis Presley reportedly staged his own death, for personal reasons. One of the most famous but oft overlooked conspiracy theories of the 1960s involves another “faked” death — that of one of the most popular musicians not only of that era but of all time, Paul McCartney of the Beatles.

After a brief overview of several famous conspiracies and the reasons for their popularity, the latter theory is discussed in depth. In 1967, popular culture created the conspiracy of the accidental death of the artist in an automobile crash. Extensive retrospective and prospective analysis of the group’s musical output (including LP albums, packaging materials, movies, etc.) uncovered numerous “clues” supporting the veracity of the theory, some dating back to the 1965 *Rubber Soul*. These, combined with the band’s sudden cessation of public concert performances in

August of 1966, were taken as undeniable evidence of a conspiracy. More familiar specific clues include:

I buried Paul (*Strawberry Fields Forever*)

Paul is a dead man - miss him, miss him, miss him (*The Beatles*)

Turn me on, dead man (*The Beatles*)

Famed attorney F. Lee Bailey presented the case on a television special in 1969, followed shortly by a *Life* magazine article in which the artist denied everything. Despite this disavowal, musicologists have documented evidence through the Beatles’ final album *Let it Be* in 1970 — but the story did not stop there. Popular media referring to the incident have also included the graphic novel *Batman* (1970), the hit television show *The Simpsons* (1990), and even the “victim” himself in his live concert album *Paul is Live* (1993).

Indeed, the surviving band members apparently included subtle additional clues in the Beatles 1995 reunion’s *Free as a Bird* music video. Additional references have been included as recently as 2009 in Italy.

Numerous visual and auditory clues are presented for interpretation by the audience to evaluate the existence and veracity of the clues, in order to develop a deeper understanding of this conspiracy theory from the standpoint of marketing and significant contemporaneous societal factors. Finally, the popularity and pervasiveness of this legend will serve as a model for understanding how such a seemingly impossible theory can gain traction and itself become an enduring part of the social lexicon.

Conspiracy Theory, Music, Beatles

BS2 The Boxer, the Priest, the Ex-Cop, and the IRA

Ronald Brunelli*, 100 Elizabeth Blackwell Street, Syracuse, NY 13210; and Robert Stoppacher, MD, 100 Elizabeth Blackwell Street, Syracuse, NY 13210

After attending this presentation, attendees will become familiar with an international cast of characters involved in a multi-million dollar heist of a Brinks® depot, the subsequent death and forensic identification of one of the planners, and steps taken to return him to his native land.

This presentation will impact the forensic science community by providing an interesting case of lessons learned from the discovery and identity of a foreign-born individual. Specifically, it will highlight issues related to an accurate anthropologic evaluation of dismembered, skeletonized remains, illustrate the need for persistence and interagency cooperation in the investigation of unidentified human remains, and underscore the value of the National Missing and Unidentified Persons System (NamUs) and Combined DNS Index System (CODIS) in the identification of human remains.

In January 1993, a group of masked gunmen, one with

an Irish brogue, held up a Brinks® depot in Rochester, NY, and escaped with \$7.4 million dollars. The only security guard at the depot was a retired Rochester, NY, police officer (the ex-cop) who was known to have ties to the Irish Republican Army (IRA) as he had reportedly helped smuggle an ex-IRA member into the United States and had also befriended a suspected gun runner for the IRA, a New York City priest (the priest). A third individual, a retired boxer from Ireland (the boxer), came to New York City in the 1970s and ended up working in an illegal casino where he met the other players. Although he knew of the plan, the boxer was reportedly cut out of the heist as he was deemed to be too irrational.

In November 1993, the Federal Bureau of Investigation (FBI) and Rochester Police Department followed the trail of the money to New York City. There, the priest and ex-IRA member were found in an apartment with \$2 million. Both were arrested and convicted while the ex-cop was acquitted. It is speculated that the remaining \$5.4 million went to the IRA.

Then, in August 1995, the boxer, having been cut out of the deal, attempted to get what he felt was his share of the prize. He borrowed a car from his friend to travel from New York City to Rochester. The car was later found in an Applebee's® restaurant parking lot in the Rochester suburb of Greece, NY, but the boxer, Joseph Ronnie Gibbons, never returned.

In June 1999, a human foot with a New Balance® sneaker was found on the east shore of Lake Ontario, in the town of Cape Vincent, NY. This was sent to the Onondaga County Medical Examiner's Office (OCMEO) and the manufacture date of the sneaker was established. Over a year later, a torso clad in blue shorts with a New York Athletic Club logo was found on an island east of the shoreline where the foot had been found. A local radiologist determined the torso to be that of a female. Two years later, the torso was brought to the OCMEO and determined to be male by both radiologic and anthropologic examination and DNA testing by a private laboratory confirmed the torso and foot were from the same individual.

Through ongoing forensic investigation and contact with multiple law enforcement agencies, the case was entered into NamUS and the remains were tentatively identified as those of the boxer. Subsequent DNA samples were recovered from the exhumed torso and family members in New York City and the United Kingdom, all of which were entered into CODIS, confirming the remains to be those of Joseph Ronnie Gibbons.

Although it took more than ten years to identify the boxer, the family was able to have the remains returned to England for proper burial. To this date, no one has been charged with Gibbons' murder; however, police are continuing their investigation.

Cold Case, Unidentified Remains, NamUs

BS3 Electrical Death, Injury, Arc-Flash, and Lightning Investigation Methods

Helmut G. Brosz, PEng, BAsC, Brosz Forensic Services, 64 Bullock Drive, Markham, ON L3P 3P2, CANADA*

The goal of this presentation is to provide an overview of the key steps and investigative methods used when dealing with electrical death and injury.

This presentation will impact the forensic science community by improving the ability

of Authorities Having Jurisdiction (AHJ), including medical examiners, police and fire departments, insurers, lawyers, prosecutors, and engineers, to investigate and document complex scenes where an electrical incident has taken place. Attendee knowledge of electrical phenomena is expected to increase.

AHJ attending to an accident scene or homicide scene

involving possible electrical phenomena sometimes have difficulty discerning between natural death and death involving electricity, including lightning. There are on average, between 500 and 1,100 electrocutions per year in the United States and Canada including suicide, auto-erotic, accidental, homicide, etc. There are also about ten times as many arc-flash burn injuries and more than ten times as many nonfatal electric shocks. Some minor shock situations cause falls or reactions which can lead to death and injury. Low-voltage electrocutions often leave no visible marks on the deceased. High-voltage electrocutions mostly leave distinctive marks and devastating injuries.

Lightning deaths may or may not leave readily identifiable marks. Arc-flash burns may sometimes obscure contact entry and exit marks. Scene photos, hospital admission notes and sketches, autopsy photos, and reports are important data to be created, then assembled, studied, and analyzed.

Review and study of applicable standards and codes can assist the AHJ or investigator. For example, electrical utility contact accidents involve the National Electrical Safety Code (NESC) or an equivalent or similar state code such as General Orders 95 and 165 and 128 as in California. The other significant regulations are OSHA 26 CFR 1910 and 26 CFR 1926. OSHA regulations also rely on NFPA 70E to a significant degree.

The National Electrical Code (NEC) which applies to residential, commercial, and industrial electrical construction is the governing rule book for many states. Cities such as New York City and Chicago, for example, have created their own rules in this regard.

Other standards pertain to the electrical products such as Underwriters Laboratory/Canadian Standards Association (UL/CSA). International Electrotechnical Commission (IEC) may also apply and will be discussed briefly. This presentation will provide a brief overview from a forensic engineering perspective of the physics and biology of electrocution and shock to humans and animals.

Overhead downed power line contacts with humans can lead to electrocution. Power line contacts with vegetation can lead to wildfires and other problems. This breakfast will be supported by presentations of cases, examples, and artifacts. Laboratory replication of electrical incidents will also be discussed.

Electrocution, Arc-Flash, Shock

BS4 No Shelter From the Storm: The Heroes and Villains of Hurricane Katrina

Steven B. Karch, MD, PO Box 5139, Berkeley, CA 94705-0139*

After attending this presentation, attendees will: (1) view previously unseen evidence of the storm damage; (2) hear a critique of the various types and relative reliability of expert testimony involved; (3) hear a review of the toxicology issues encountered in the disaster settings; and, (4) explore recurrent legal issues and have the opportunity to ask questions.

This presentation will impact the forensic science community by increasing awareness of death investigation in a post-disaster setting.

Katrina was one of the greatest natural disasters ever to strike the United States. By the evening of Monday, August 29, 2005, more than 80 percent of New Orleans lay underwater. By the time the water had receded and a body count attempted, more than 1,600 lives had been lost, 81,000 individual businesses had been destroyed, and more than 200,000 homes had been severely damaged or washed away. The federal government was nowhere to be seen except during the now infamous over-flight of Air Force One.

Two weeks after the storm, 45 decomposing bodies were found in the hulk that had been Memorial Hospital (11 of those who died were in the hospital's morgue prior to the storm). Of the 34 remaining patient deaths, 24 were from the Lifecare (acute care) specialty hospital within Memorial, which swelled to a patient population of over 50 by the last-minute transfer of 20 Lifecare patients from another hurricane-threatened area.

Ironically, many of the Lifecare patients, who were later the focus of a criminal investigation, had been transferred to Memorial for their own safety. Lifecare's medical director evacuated, leaving his patients in the care of others. The misery of the conditions under which they died can never truly be described. Memorial was without electricity, had no running water, no air conditioning, inadequate supplies, with an inside temperature mostly in the 100- to 110-degree range. Toilets were overflowing, bodies were decomposing, and the stench was overbearing.

Dr. Anna Pou, who was about to become an unwilling participant in a media circus, stayed through it all, and ended up being accused of murder for her efforts; she and two colleagues were charged with having systematically euthanized many patients. When a special Grand Jury rejected the findings of the then — Louisiana Attorney General Charles Foti and refused to return an indictment, Dr. Pou was still left to face an endless sea of malpractice litigations and media scrutiny. The cases against Pou were largely based on sealed records that were faxed from the Attorney General's office to CNN and a local newspaper. All of these cases have now been dismissed. No court ever ruled against Dr. Pou, and the criminal charges against her and two nurses who were also arrested and accused have now been expunged. While the deaths happened, the murders did not.

There is no statute of limitations on murder and another District Attorney might try to bring charges, but the statute of limitations for civil litigation has expired. No more civil actions will be brought against Pou or her colleagues. For that reason, attendees will hear a documented account of the damage wrought by the storm, and the legal fallout that followed — at least so far as one member of the defense team was able to determine. The records of the investigation remain sealed, but that has not kept the details from being selectively released to the media, apparently by Foti's investigators, an ethical breach at a minimum and, in reality, a probable crime in and of itself.

Even though it was never proven that Dr. Pou had harmed anyone, her career and the careers of the two nurses were all tarnished and their lives disrupted in ways that cannot be imagined while the threat of criminal charges and civil suits existed. Some less-than-scrupulous writers and television producers took advantage of the fact, knowing full well that the accused could neither speak nor defend herself in any way. Indeed, a new book based on documents leaked from the government was released in September 2013.

If it were possible to identify just one single feature that made Hurricane Katrina so devastating, most agree it was the complete and utter failure of the federal government to offer aid to the distressed and dying, and to organize a comprehensive, efficient evacuation of those in need. There are lessons to be learned from Katrina about disasters and about how to manage them. Perhaps more importantly, we have learned something about the way institutions and individuals respond in times of great hazard. There are even more important lessons to be learned by the forensics community and the courts. They are neither encouraging nor inspiring.

Hurricane Katrina, Postmortem Toxicology, Euthanasia

BS5 Thomas Krauss Memorial Bitemark Breakfast — The Role of an FBI Forensic Odontologist/Special Agent in Federal Crime Scenes

Scott H. Hahn, DMD, FBI Miami, ERT, 16320 NW 2nd Avenue, North Miami Beach, FL 33169*

After attending this presentation, attendees will gain an understanding of the jurisdictional requirements and limitations regarding federal crime scenes (and/or federal referrals) wherein the Federal Bureau of Investigation (FBI) employs a Special Agent forensic odontologist to assist in the identification of human remains. Attendees will also gain an appreciation for the concept of the FBI having the capabilities to forward deploy a Special Agent forensic odontologist to exigent and/or unique scenes to perform identifications or eliminations of unidentified human remains of interest to U.S. federal law enforcement.

This presentation will impact the forensic science community by further validating the odontological examination and comparison process as being a universally accepted form of human remains identification. Further, the utilization of the "front-line" forensic dentist, as outlined in the presentation, may prompt other entities involved in the identification of human remains to consider similar type "go-teams" (U.S. Military Armed Forces Institute of Pathology (AFIP) model, FBI Evidence Response Team (ERT) model) for smaller, non-mass casualty events.

Historically, the FBI relied on the services of the Smithsonian Institute Department of Anthropology and the Armed Forces Institute of Pathology Odontologists to assist the Bureau in the identification of human remains recovered during FBI investigations (or referred to the FBI by other law enforcement entities). Within the last several years, the FBI Laboratory in Quantico, VA, initiated an in-house anthropology department staffed by two full-time, board-certified anthropologists. Likewise, the FBI Evidence Response Team Unit (an operational entity of the FBI Laboratory) paved the way for an FBI board-certified odontologist Special Agent to address the forensic dental aspects of FBI investigations.

The majority of FBI cases requiring a forensic odontologist are referred to ABFO members in the geographic area of the investigation. However, when the rapid deployment of forensic dental assets is required overseas and/or in sensitive investigations and/or dangerous locales, a Special Agent odontologist is uniquely qualified for those special circumstances. The deployment usually involves several ERT personnel in addition to the odontologist, such as Special Agent photographers, evidence experts, and force protection elements.

Recent FBI odontology cases have involved the deployment to other countries to identify U.S. victims of violent crimes and/or acts of terrorism. In some cases, the host nation will not relinquish the U.S. citizen's remains without a scientific identification, and forensic odontology has been a universally accepted methodology to meet that requirement. In other FBI cases, the Special Agent odontologist has been deployed to perform *in situ* identifications and/or eliminations in less-than-permissive environments.

The terminal learning objective of this presentation is that forensic odontology, whether practiced in a Medical Examiner's Office or the jungles of a South American country, is very much a universally accepted form of scientific identification of human remains, and the basic tenets that are employed by all practicing forensic dentists work equally well in either environment.

FBI, Odontology, Federal Crime



LUNCHEON SEMINARS



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L1 Deadlier Than the Male

Ann Rule, BA, Rule Enterprises, Inc, PO Box 98446, Seattle, WA 98198*

After attending this presentation, attendees will learn how forensic evidence played a large part in tracking and trapping women who were not nearly as clever as they thought they were.

This presentation will impact the forensic science community by bringing to light case

studies of female killers in the U.S. and how, in these cases, they are “Deadlier than the Male.”

Since time immemorial, women have been viewed as the weaker sex, the gentler sex. The percentage of females responsible for violent crimes has been far less than males. In recent years; however, we are seeing more women who are sadistic sociopaths. I have yet to research a woman who is a serial killer in the accepted sense, however. Women are seldom “hands-on” murderers, and they tend to kill people they know, relatives and those who trust them, whereas male serial killers most often kill stranger-to-stranger, making them more difficult to identify.

The female of our species most often employs poison — be it ant killer, anti-freeze, arsenic — or some other method. Two “ladies” in California tried everything but a black widow in a blueberry pie to more common poisons. Women’s motivation isn’t murder as part of a sexual attack or murder for its own sake; women kill for love (*VERY broadly defined*) or for money. They also plan their homicides with great care and are willing to wait until the time is right.

Often, women don’t want to get their hands bloody, literally or figuratively, so they seduce males to carry out their plans.

Having written about a number of females who committed murder — or caused murder to be committed, several cases will be discussed: Diane Downs, who shot her own children (a blueprint used by Susan Smith a few years later); Dr. Debora Green, who murdered *her* children by fire; Patricia Radcliffe Taylor Allanson, who murdered by “proxy”; Liysa Northon, who used heavy drugs in her cooking, near-drowning, and, finally, a bullet right between the eyes to kill her husband; and, Teresa Gaethe who crept into her estranged husband’s bedroom and shot him as he lay in his waterbed.

The seminar will end with a discussion of my current book, *Practice to Deceive*, about Peggy Sue Stackhouse Harris Thomas Allen, one of the most manipulative women I have ever researched.

The presentation will include a number of photos as I move from the rare murderesses in history to my current book, *Practice to Deceive*.

Women, Serial, Killers

L2 Medical Ethics Simplified — A Model for Ethical Conduct for the Forensic Science Practitioner

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After attending this presentation, attendees will be familiar with a brief history of medical ethics in the U.S., dating to 1803. The attendee will also learn a simplified ethical code as a mnemonic for continued practical application.

This presentation will impact the forensic science community by imparting that by learning the history of modern medical ethics in the U.S. and by considering a medical model as a possible structure for other forensic disciplines, a simplified paradigm modeled on the classical seven deadly sins and seven cardinal virtues will facilitate the practitioner’s pursuit of a virtuous career.

The National Academy of Sciences 2009 Report cited a critical need for the establishment of ethical standards for conduct¹

Standards and codes of ethics exist in some fields, and there are some functioning certification and accreditation programs, but none are mandatory. In short, oversight and enforcement of operating standards, certification, accreditation, and ethics are lacking in most local jurisdictions.

... a national code of ethics [should be established] for all forensic science disciplines...individual societies (are encouraged) to incorporate this national code as part of their professional code of ethics. Additionally,...mechanisms of enforcement for those forensic scientists who commit serious ethical violations (should be explored). Such a code could be enforced through a certification process for forensic scientists.

...Congress can promote “best practices” and strong educational, certification, accreditation, ethics, and oversight programs in the states by offering funds that are contingent on meeting appropriate standards of conduct.

A cultural foundation must be implemented early in order to maximize acceptance by those directly involved. As noted in the NAS Report, ethics training should be no less a part of the core forensic science curriculum than the natural sciences. As the practice of medicine has many such elements already implemented, the discipline and history might serve as a useful model for other forensic disciplines in the ongoing efforts for improvement, ultimately leading to a workable ethics code.

Percival’s Medical Ethics, released in 1803, became the basis for the original standards of ethical conduct for the American Medical Association and to some extent eventually the entire medical system in the United States.² In this treatise, Percival used a case-study approach, familiar to those in the field. He interspersed his rules with memorable insights and quotes, illustrating a greater practical understanding of the issue of ethics *in context*, rather than merely a dry recitation of enumerated commandments. Although

the work is seriously dated, it was created for the purposes of addressing specific needs at a particular moment in time in a particular location. The strength of the work remains the relevance to those who needed the information contained therein.

Based on Percival's model, a greatly simplified core ethical system is presented — that of the classical "seven deadly sins" made famous by Dante (lust, envy, greed, sloth, gluttony, anger, and pride). Utilizing a case-study approach demonstrating each of these transgressions in a forensic setting, as well as the countervailing cardinal virtues (chastity, kindness, charity, diligence, abstinence, patience, and humility), a simplified ethics mnemonic is suggested. Understanding of precepts requires not only exposure to the concept but acceptance. The hope is that forensic service providers will be inclined to utilize the simple seven deadly sins as an aid to continued pursuit of an ethical career.

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2. Percival's Medical Ethics. The moral philosophy of an 18th-century English gentleman. Pellegrino ED. Journal, Arch Intern Med.1986 Nov; 146 (11):2265-9

Medicine, Ethics, NAS Report



WORKSHOPS



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W1 Designer Drug Detection in Forensic Toxicology: From Basics to Brilliant!

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After attending this presentation, attendees will better understand the analytical challenges associated with designer drug detection. This presentation will also assist toxicologists in expanding their repertoire of testing in a routine setting.

This presentation will impact the forensic science community by improving routine forensic toxicology investigations and, more specifically, improving and expanding current analytical capabilities so that laboratories can identify these emerging substances during routine testing.

Forensic toxicology laboratories are faced with an expanding array of new and emerging drugs. Synthetic cathinones and canabimimetics represent two important classes of designer drugs that pose a formidable challenge to busy laboratories that perform routine testing in a wide array of forensic investigations. This workshop will address some of the challenges associated with detection and provide information that will improve toxicological investigations involving these substances.

Many toxicology laboratories in the United States still lack the necessary resources and instrumentation to perform highly specialized testing for the growing number of synthetic drugs that are being used. Nevertheless, even laboratories with limited resources must implement techniques to identify cases where cathinones and canabimimetics are present in biological samples. Failure to do so could have serious consequences in antemortem and postmortem forensic toxicology investigations. This workshop will focus on a variety of strategies to improve the scope of testing for a wide array of laboratories with varying resources.

The characteristics of these two classes of drug will be reviewed in terms of their overall chemistry, classification, and the properties that must be taken into consideration when developing new methods for extraction and instrumental analysis. Incorporation of designer drug testing into routine casework will be discussed from the perspective of the laboratory director. Challenges and strategies to maximize success and rely on existing resources will be emphasized. The scope and limitations of what can be accomplished with and without highly specialized instrumentation will be explored so that laboratories can make informed decisions when evaluating cases that may potentially involve these substances.

Techniques for screening and confirmation by immunoassay, Gas Chromatography/Mass Spectrometry (GC/MS) and Liquid Chromatography-Tandem Mass Spectrometry (LC/

MS/MS) will be presented. The advantages, disadvantages, and most appropriate techniques will be discussed for each designer drug class. Analytical testing and methodology for comprehensive testing will be presented. These techniques will be discussed within the context of interpretive issues, limitations, and challenges. Stability and other factors that may influence quantitative analysis and subsequent toxicological interpretation will also be discussed.

Workshop attendees will be exposed to a variety of screening and confirmatory methodologies that will help them improve their analytical capability in a way that works for their organization, resources, and overall mission. Incorporating new and improved techniques to detect these designer drugs in biological samples will improve the routine practice of forensic toxicology including death investigation, impaired driving, and other toxicological investigations where synthetic cathinones and canabimimetics may be encountered.

Designer Drugs, Cathinones, Cannabimimetics

W2 Advanced Forensic Pathology and Forensic Radiology Techniques in the Assessment of Cervical Spine and Craniocervical Junction Trauma

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After attending this session, attendees will understand: (1) the role of radiography and advanced radiologic imaging techniques in a medical examiner setting; (2) the anatomy of the cervical spine and craniocervical junction; (3) advanced autopsy techniques that allow for the assessment of the cervical spine and craniocervical junction, including intrinsic anatomy (neurovascular structures); (4) the value and limitations of radiography of the cervical spine and skull base; (5) the value of postmortem computed tomography in the assessment of the cervical spine and craniocervical junction; and, (6) the value of postmortem magnetic resonance imaging in the assessment of the cervical spine and craniocervical junction.

This presentation will impact the forensic science community by teaching advanced autopsy and radiologic techniques that facilitate documentation and diagnosis of trauma and other pathologies in the cervical spine and craniocervical junction.

Cervical spine and Craniocervical Junction (CCJ) trauma is frequently encountered by forensic pathologists and medical

examiners. Whether occurring alone or in combination with other more complex injuries, trauma in these regions is frequently cited as causing or contributing to death. However, cervical spine and CCJ trauma is notoriously difficult to detect, thoroughly document, and accurately diagnose. While so-called "typical" anterior and posterior neck dissections can address these objectives, more advanced methods of evaluation are often required. Examples include *in situ* evaluation of ligamentous and chondro-osseous structures (including the odontoid process and its supporting ligaments) and *ex situ* evaluation of all intrinsic structures including the neurovasculature and spinal cord.

Radiography has long been a tool in the arsenal of the forensic pathologist and can be used in the assessment of cervical spine and CCJ trauma; however, correct interpretation is predicated upon the technical quality of the radiographs and the knowledge base of the interpreting pathologist. In recent years, Postmortem Computed Tomography (PMCT) and Postmortem Magnetic Resonance (PMMR) imaging have been utilized with greater frequency in medicolegal death investigations. Both PMCT and PMMR have complementary roles in the assessment of the cervical spine and CCJ. In some circumstances, data obtained from either PMCT or PMMR obviate the need for advanced dissections of the cervical spine and CCJ. In the appropriate historical circumstances, information obtained from advanced radiologic imaging studies may supplant autopsy altogether when combined with thorough external examination and appropriate ancillary studies. However, a combination of both PMCT and PMMR would be required to produce a similar level of detail and accuracy as would be obtained from advanced autopsy dissection techniques.

Cervical Spine, Forensic Radiology, Autopsy Dissections

W3 Development of Emerging DNA Technologies for Identification: Expanding the Capabilities of a Missing Persons Laboratory

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After attending this presentation, attendees will learn about new and emerging forensic DNA technologies and their application in missing persons efforts.

This presentation will impact the forensic science community by detailing the various steps involved in the development, optimization, and validation of new procedures in a forensic laboratory and how the implementation of these methods into forensic casework impacts identification capabilities.

The Armed Forces DNA Identification Laboratory (AFDIL) annually processes approximately 1,300 skeletal specimens from past military conflicts. In these decades-old cases, identification is primarily achieved through mitochondrial DNA analysis, but this is often limited by common haplotypes and unavailable maternal references. To address these limitations, the AFDIL continuously strives to develop, optimize, and validate new techniques to assist with the re-association of commingled remains and support identifications in missing persons cases. This workshop will focus on various projects that have already made an impact on day-to-day casework at the AFDIL, as well as investigations of novel technologies that promise to benefit the on-going efforts to identify the nearly 83,000 U.S. service members still missing.

AFDIL annually processes approximately 1,300 skeletal specimens from past military conflicts. In these decades-old cases, identification is primarily achieved through mitochondrial DNA (mtDNA) analysis due to its high copy number and maternal inheritance. However, the forensic utility of mtDNA data is limited by uniparental inheritance and lack of recombination, which can sometimes result in a low power of discrimination due to common haplotypes. This testing also requires either direct or maternal references for comparison, and in some cases appropriate references are unavailable. There is a growing need to utilize reference samples from a relative that may be several generations removed, and data from additional DNA markers in the nuclear genome could benefit identification efforts. However, the poor quality and limited quantity of nuclear DNA present in degraded skeletal remains has historically restricted the use of Short Tandem Repeat (STR) data in such cases. Additionally, the postmortem conditions and treatment of some specimens can damage the DNA so severely that no genetic data can be generated. To address these limitations, the AFDIL continuously strives to develop, optimize, and validate new techniques to assist with the re-association of commingled remains and support identifications in missing persons cases.

Recent work has focused on improving current methods, expanding the type and amount of genetic information obtained, streamlining current workflows through automation, and evaluating promising new technologies. To begin, the validation of a non-organic extraction procedure for low-quality skeletal specimens has demonstrated improved extract recovery and purity by combining full decalcification with silica-based purification. The recovery of nuclear DNA data has recently been made possible through the validation and implementation of a modified amplification strategy using a commercial Y-chromosomal STR kit. This ability to obtain nuclear information allows the relevant pool of family reference samples to be expanded, a goal that is also being pursued through other novel approaches such as the use of X-chromosomal and non-Combined DNA Index System (CODIS) autosomal STRs. Access to this information continues to be a challenge, and recent efforts have focused on the evaluation of novel DNA sequencing methods known collectively as Next Generation Sequencing (NGS). NGS techniques hold the potential to aid in the analysis of the most challenging samples through short read lengths that are able to accommodate highly degraded and/or damaged samples and massively parallel sequencing reads covering the region of interest hundreds to thousands of times. Additionally, multiple samples can be processed across multiple marker systems at the same time, potentially decreasing both the cost and time required for analysis. This benefit of cost and time savings is also the goal of other new methods implemented at AFDIL. The automation of high throughput reference sample sequencing as well as the implementation of a species identification assay to eliminate fruitless processing of non-human specimens have both served to reduce the time and cost of results.

With the introduction of new and enhanced techniques, there are forensic-specific implications that must be considered and addressed in the validation prior to implementation. This workshop will discuss these considerations as well as present case examples that demonstrate how the novel methods have improved success rates and contributed to on-going efforts to identify the nearly 83,000 U.S. service members still missing.

The opinions or assertions presented herein are the private views of the author(s) and should not be construed as official or as reflecting the views of the Department of Defense, its branches, the U.S. Army Medical Research and Materiel Command, or the Armed Forces Medical Examiner System.

Skeletal Remains, Missing Persons, Emerging DNA Technologies

W4 Virtual Evidence of Human Behavior: Technology as Co-Conspirator

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After attending this presentation, attendees will be able to: (1) list three technologies impacting human behavior; (2) give three examples of cyber-interactions changing the practice of medicine; (3) describe hybrid crime assessment; and, (4) demonstrate familiarity with one published classification scheme of four offender subtypes.

This presentation will impact the forensic science community by promoting multidisciplinary discourse and learning.

Advances in digital and multimedia technology are significantly impacting human behaviors, social interactions, and vice versa. This two-way interchange between technology and behavior blurs the focus on technology, as opposed to technology being the lens for better understanding human behavior. The combined areas of expertise span primarily two American Academy of Forensic Sciences (AAFS) sections: (1) Psychiatry and Behavioral Science; and, (2) Digital and Multimedia Sciences. However, this presentation explicitly assumes only basic familiarity with discipline-specific knowledge and terminology.

The internet has, so far, generally enabled participants in on-line interactions to determine their level of identification. This significantly affects behavior, rendering some more likely to say or do things that they would not in their everyday lives off-line. Some of these exchanges represent beliefs generally not acted on or are simply intended to provoke a response from others. This interplay of identity, anonymity, and behavior creates unique challenges when interpreting digital evidence. It can be difficult to extrapolate one's behavior off-line (in "real" life) from one's cyber behavior, even in circumstances where one's identity is adequately verifiable. This interplay also leads to novel forms of harassment such as doxing and extensions of pre-existing harassment such as bullying.

On occasion, self-expression through digital media becomes the focus of clinical attention. One's responses to being bullied may be aired out in the public cyberdomain. Modern-day mental illnesses may present in the form of suicide notes published on-line, and acts of self-injury or even suicide completion videos available on the internet. Some websites promote behaviors symptomatic of conditions such as anorexia nervosa.

Computers not only affect human behavior, but they can be rich sources of digital evidence reflecting the thinking and activities of the real world. Electronic mail and on-line social networks can serve as evidence analogous to the narratives of eye witnesses. Unlike considerations of recall bias in eye-witness testimony, digital evidence has higher reproducibility. However, manipulation and interpretation of digital data structures introduce new concerns with respect to the accurate truth, but also present novel applications: (1) tele-medicine is delivering increasing care to historically underserved populations. Behavioral emergencies may arise, e.g., a patient barricaded in a hotel room with weapons. In these situations, basic information (such as the patient's physical location) might be clinically challenging to ascertain; (2) state medical boards may use cyberdata as evidence of doctors' sexual relationships with patients or of breaches of patient confidentiality; and, (3) lastly, crime scene interpretation exemplifies another area of novel application. "Hybrid crimes" are physical crimes joined with

a significant digital component, such as internet or social media, which create a single, hybrid crime scene where the common thread is the criminal. In hybrid crimes, digital evidence augments the understanding and interpretation of the physical crime scene. Behavior in one venue—cyber, for example—can inform the analysis of a crime in its entirety.

Narrative approaches used in forensic psychiatry (particularly when human dramas invoke larger discussions on morality) can inform the behavioral analysis of crime scenes. Narratology in the interpretation of crime scenes is based upon the premise that every crime has an associated story, with a plot, characters, venues, etc. The story begins some time before the crime and, as electronic events transpire, the cyber world captures and preserves at least basic data such as date and time. Technologically-assisted temporal association of events with the crime and with each other can significantly enhance both the narrative and one's ability to understand the story behind the crime.

One approach to hybrid crime assessment incorporates previous work in describing four distinct subtypes of criminals: power-assertive; power-reassurance; anger-retaliatory; and anger-excitation or sadism. This approach, and its potential application as an investigatory tool for forensic psychiatrists and other behavioral scientists, will be discussed. Other directions for future investigation, including cybercrime as a potential predictor of non-cybercrime (such as rape or assault), will also be discussed.

Human Behavior, Advanced Technology, Internet

W5 Forensic Microbiology: Where Do We Begin?

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After attending this workshop, attendees will have an understanding of microorganisms and their roles in forensic science.

This presentation will impact the forensic science community by providing current knowledge and techniques for analyzing microbial communities.

The application of microbiology to investigations of decomposing human remains has now become a major area of research within the forensic community. Presently, researchers are deciphering bacterial communities associated with human remains in order to estimate: (1) postmortem interval; (2) geographic origins based on microbial biogeography of living human populations; and, (3) the use of narcotics or poisons prior to death.

Upon completion of this workshop, the participants should be able to collect, preserve, and ship microbial samples to appropriate experts for analysis. Participants will be able to assist employers with the development of laboratory facilities and personnel necessary for analyzing forensic microbial samples in-

house. Participants will gain an appreciation for the application of microbial forensics within their facilities to estimate when a person died, where they were from, and if they consumed or were treated with chemicals of human importance.

This workshop will stimulate discussion among international, federal, state, and private laboratories on topics germane to forensic microbiology, which is revolutionizing how microbes are viewed in medicolegal death investigations. Currently, standards are not in place for such work and this workshop could result in the establishment of a working group to develop such standards.

Microorganisms, Forensic Microbiology, Investigations

W6 Staged Crime Scenes: Crime Scene Clues to Suspect Misdirection of the Investigation

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After attending this presentation, attendees will learn three new categories of staged scenes, with case examples from different types of crimes where staging is prevalent. Additionally, attendees will learn the “red flags” or common findings in staged scenes.

This presentation will impact the forensic science community by introducing distinct categories of staging based on the intent of the offender’s scene alteration. Ultimately, applying these categories may help in the quick recognition of scene alterations or staging.

In the course of their career, most detectives and forensic practitioners will come into contact with a staged crime scene, i.e., a scene that has been altered by the offender to either mislead a police investigation as to the true facts of the crime or for other reasons understood only by the offender. Staged scenes and “staging” are possible in nearly every type of criminal offense ranging from property crimes such as arson and burglary to violent crimes such as homicide, child abuse, or sexual assaults.

To better understand the dynamics and the general nature of “staging,” this work shop will introduce distinct categories of staged crime scenes based on the intent of the offender’s scene alteration. The intent of this workshop is to understand that the offender’s actions to stage a scene can actually be identified through common findings or “red flags” that are often found when scenes are altered. Further, attendees will learn that these same red flags may help in the quick recognition of scene alterations or staging and, based on new criteria, may be divided into three separate and distinct categories. These categories are herein referred to as primary, secondary, and tertiary.

The primary staged scene is intentionally altered or changed by the offender with criminal intent to misdirect a subsequent police investigation and can be further sub-categorized into two types: *ad hoc* and premeditated. The *ad hoc* subtype is staged without forethought and planning, at the spur of the moment after the event has taken place, and is generally intended to deflect attention away from the offender and the true facts of the crime. The scene is considered premeditated when the offender pre-plans the scene alterations in accordance with a preconceived scenario. Premeditated staging is often designed to focus attention onto the staging and false evidence. Ultimately, through primary staging, the offender in effect creates a false reality that in his/her mind will successfully and with criminal intent misdirect the police investigation.

Secondary staging involves the intentional alteration or

manipulation of the crime scene or victim by an offender that is unrelated to misdirecting or diverting subsequent investigations. This is really a new category of staged scenes and would include such examples as posing the victim’s body into sexually provocative positions as found in sexual homicides or other elements of what is more commonly known as “staging,” such as covering the face or body in what is often described as depersonalization. The perpetrator’s purpose in secondary staging is not to misdirect the investigation; rather it is often something that is psychologically part and parcel to the crime, such as demeaning the victim or demonstrating ultimate control over the victim.

The third category to be discussed and defined consists of noncriminal, accidental, or innocent alterations; i.e., changes to the original crime scene, generally by witnesses or family members, who find the victim and alter the scene without any criminal intent. An example would be a family member finding a loved one in an embarrassing position from an autoerotic misadventure and changing the scene to prevent embarrassment to the family. These types of alterations are better described as tertiary; and are best regarded as scene artifacts.

This workshop would have application to persons in forensic pathology, criminalistics, crime scene analysts, and criminal investigations. It introduces three new categories of staged scenes, provides case examples, and explains the “red flags” commonly encountered when confronted with a staged scene. The workshop culminates with case studies for each student to work through and identify the various “red flags” in real cases.

Primary Staging, Secondary Staging, Tertiary Staging

W7 Age Estimation of Living Children Via Skeletal and Dental Indicators

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After attending this presentation, attendees will: (1) understand the basic principles and techniques involved in age estimation of living children via radiographic analysis of skeletal and dental indicators; (2) be aware of the current literature dedicated to this topic; and, (3) be given the opportunity to practice what they have learned on a series of radiographs of known ages.

This presentation will impact the forensic science community by preparing forensic practitioners for the increasing demand by law enforcement agencies to estimate the age of living children who lack proper documentation of their birth. A comprehensive manual containing practical data dedicated to meeting this need will be provided. Considering a larger impact scope, attendees will be better prepared to estimate the age of children in general (be they living or deceased) via radiographic analysis.

With immigration on the rise, forensic practitioners are increasingly being asked to estimate the age of living children who lack proper documentation of their birth. As it stands, there is no comprehensive manual containing practical data dedicated to helping practitioners meet this demand. Thus, practitioners who are new to this request may not be aware of the methodologies and resources available. The goal of this workshop is to fill that void by reviewing the fundamental concepts involved in estimating the chronological age of living children and providing attendees with a collection of available data to enable this task.

Estimating age based on hard tissue analysis is normally accomplished via investigation of both dental and osseous

maturity. However, age estimation of the living is unique in that analysis is restricted to various types of radiological images. Thus, practitioners must be confident in their ability to interpret developmental changes demonstrated within radiographic, ultrasound, Computed Tomography (CT), and/or, Magnetic Resonance Imaging (MRI) images. This workshop will review developmental processes that take place in the dental arcade as well as each of the joint regions of the upper and lower extremities via radiographic images. Studies pertaining to each region will be introduced and discussed, highlighting the positive and negative attributes of each technique. While interpretation of all juvenile ages will be addressed, emphasis will be placed on interpreting developmental milestones that are useful in the determination of whether an individual is older or younger than popular threshold ages that are commonly pertinent to the penal system.

Age Estimation, Juvenile Development, Radiographs

W8 Utilizing Bloodstain Pattern Analysis and Forensic Pathology to Reconstruct Bloodshedding Events

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After attending this presentation, attendees will be: (1) acquainted with the basic concepts of bloodstain pattern analysis; (2) able to illustrate the correlation between injuries and bloodstain patterns; (3) able to illustrate the role of pathology findings within a reconstruction with bloodstain pattern analysis; (4) acquainted with the recommended Scientific Working Group on Bloodstain Pattern Analysis (SWGSTAIN) terminology; (5) able to illustrate how bloodstain pattern analysis can assist in selecting the most probable stain(s) from a scene or article of evidence for DNA testing; (6) able to demonstrate how, if properly analyzed, bloodstain patterns can assist in reconstructing past events; and, (7) able to provide an understanding of the limitations of bloodstain pattern analysis.

This presentation will impact the forensic science community by informing the attendees how bloodstain pattern analysis and forensic pathology work together to assist in reconstructing death scenes where bloodshed has occurred.

Description: This presentation will address the primary concepts of bloodstain pattern analysis, while providing an understanding of how the physical characteristics of bloodstains patterns can assist an analyst in reconstructing scenes of bloodshed. This workshop will acquaint attendees with the recommended SWGSTAIN terminology, which is being used in proficiency testing as well as certification programs within the United States. In addition to the theory of bloodstain pattern analysis, a number of case examples will be presented throughout the workshop to reinforce the practical aspects of this valuable forensic tool.

This workshop will provide attendees with an understanding that forensic pathology is an essential skill set used in reconstructing bloodshedding event(s) with bloodstain pattern analysis. The roles of the forensic pathologist and the bloodstain pattern analyst will be clearly explained within this workshop. Attendees will understand some of the limitations encountered by the bloodstain pattern analyst and the forensic pathologist in a forensic investigation.

This presentation will utilize a unique format featuring a forensic pathologist discussing traumatic bloodletting injuries followed by a bloodstain pattern analyst discussing the bloodstain

patterns commonly associated with these traumatic injuries. This approach will give the attendees a greater understanding of how a victim(s)' injuries relate to the bloodstain patterns identified within a death scene.

In crimes of violence, blood and bloodstain patterns are commonly encountered pieces of forensic evidence which hold a significant amount of value when thoroughly analyzed. Bloodstain pattern analysis is conducted in two phases: (1) pattern analysis; and, (2) reconstruction with bloodstain pattern analysis. In Phase 1 pattern analysis, the evaluation of the physical characteristics (size, shape, distribution, and overall physical appearance) of bloodstain patterns and the consideration of the effect of the target surface texture on the pattern(s) occur. With this information, the identification of basic pattern types (passive, spatter, and altered) is determined. In Phase 2 reconstruction, a myriad of case facts (serology, pathology, scene issues, etc.) are considered. The basic pattern type and case facts are then utilized to evaluate all possible explanations for a given stain pattern, with the final outcome resulting in a reconstruction of the blood shedding event(s). The incorporation of pathological findings is a critical part of reconstructing a bloodshedding event(s) with bloodstain pattern analysis. Pattern analysis **must** be the first step completed when reconstructing an event with bloodstain pattern analysis. With proper analysis, bloodstain patterns can assist forensic investigators in understanding what did and did not occur during a violent bloodshedding event.

Bloodstain pattern analysis is a valuable forensic tool in substantiating or negating the accounts given by an accused suspect or an eyewitness. It is important to have an understanding of wound pathology when conducting a bloodstain pattern analysis. As with all forensic "tools," bloodstain pattern analysis has its own inherent limitations, which will be emphasized and discussed within this workshop. When used appropriately, bloodstain pattern analysis has and continues to be a valuable tool in reconstructing past bloodshedding events by substantiating or negating these accounts.

In addition, bloodstain pattern analysis can assist the DNA analysts in selecting the most probable samples. In the course of casework, cases are continually seen where valuable resources are wasted on samples with minimal or no real forensic value to a case. Throughout this workshop, an emphasis will be placed upon using bloodstain pattern analysis to select those samples having the greatest forensic value. Developing this skill set is especially crucial in an era of increased caseloads and decreased financial resources.

This presentation is designed for forensic practitioners whose work requires them to document, sample, or analyze bloodstains in their forensic casework. With the knowledge attained through this workshop, the attendees should be better qualified to decide whether or not to pursue additional training in bloodstain pattern analysis. It should be understood that this presentation will be addressing the analysis of the physical characteristics of bloodstain patterns and not forensic biology or DNA.

Bloodstain Pattern Analysis, Forensic Pathology, SWGSTAIN

W9 Multimedia Authentication: Searching for Truth in the Digital World

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After attending this workshop, attendees will: (1) become familiar with proper media retrieval; (2) understand criteria used for

media authentication; and, (3) understand how to work with these factors in a forensic framework.

This presentation will impact the forensic science community by: (1) explaining the history of media forgery; (2) demonstrating an authentication investigation framework; and, (3) providing the tools to combat multimedia forgery.

Media authentication has become more important than ever before. Often a crucial element discovered during investigations, digital media has become a commonly contested form of evidence. Even the trustworthiness of press photography has become questionable due to manipulation. With the proliferation of digital media manipulation tools, media manipulation is a dangerous reality in the modern digital society.

Digital media authentication is a growing field of research that seeks to determine the validity of digital multimedia by investigating known signatures within a file's data combined with signal analysis of coding and compression effects on audio or image data. This presentation will discuss the media authentication process providing the user with methods of authenticating both image and audio. It will also demonstrate the incorporation of multiple tools and techniques into unified frameworks appropriate in forensic examinations where reducing examiner bias and error is crucial.

This presentation is a two-part, full-day workshop covering image authentication analysis in the morning and audio authentication analysis in the afternoon. The goal of this workshop is to provide an overall view of conducting comprehensive examinations which rely on the results of multiple analyses to inform an ultimate finding or opinion. First to be covered is a multi-faceted image authentication framework, focusing on different aspects of image creation to determine both source and authenticity. Global Analysis investigates the validity of an image as whole while Local Analysis determines pixel level manipulation. Social Media Image Attribution will trace images even after they have been processed through social media.

In the second section of the workshop, an audio authentication framework will be presented. This framework will combine both Container and Content Analysis to determine authenticity of the recording as well as the purported source. Audio Container Analysis exploits characteristics of the multimedia file format which can be used to establish media provenance. Content Analysis will cover both Global and Local analysis techniques including classical spectral analysis, butt splice detection, DC offset, Electrical Network Frequency (ENF) analysis, and compression analysis.

Media Forensics, Audio Analysis, Video/Image Analysis

W10 Bias in Forensics — Examining the Sources and Impacts of Bias on Perceptual and Cognitive Judgments Made by Forensic Experts, Strategies for Excluding or Impeaching Expert Testimony Tainted by Bias, and Proposed Solutions for Minimizing or Inhibiting Biasing Influences

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After attending this presentation, attendees will be much better acquainted with the different types of bias that can influence the outcome of forensic investigations. Attendees will learn about classic psychological research studies and real-life case histories that illustrate how bias can improperly sway the perceptual and cognitive judgments of forensic examiners and produce faulty conclusions, even absent malicious intent. Attendees will also learn about practices that should be avoided and followed in order to minimize potential biasing influences, and means by which judges can exclude and lawyers can impeach expert evidence tainted by bias.

This presentation will impact the forensic science community by clearly demonstrating how various types of bias can adversely impact a forensic examiner's visual perception and decision making. Understanding the sources of bias and learning how to limit or minimize their influence is essential for improving the reliability and accuracy of decisions made by forensic experts. In our adversarial judicial system, trial lawyers need this knowledge in order to effectively expose potential biasing influences when cross-examining an opposing forensic expert, and judges need it to be more mindful of the critical role that bias plays in producing miscarriages of criminal and civil justice. All forensic scientists and laboratory directors must be keenly aware of the potential for bias and the types of internal procedures and protocols that can and should be implemented to minimize the impact of bias in forensic investigations and casework.

In this workshop, a multidisciplinary faculty of distinguished psychologists, lawyers, and forensic scientists will provide attendees with a clear picture and concrete examples of how and why bias affects the outcome of forensic investigations. Attendees will learn about the various experimental research studies that reveal the susceptibility of investigations to the prospect of psychological error due to cognitive and motivational factors, including confirmatory biases, dynamic influences, group membership, role conflict, and escalation of commitment. The uniquely persuasive power of confessions – even false confessions that are recanted and contradicted by other evidence – will be discussed in the context of basic and forensic psychology research which indicates that confessions corrupt lay witnesses and forensic science examiners across a range of domains, thereby increasing the risk of wrongful convictions.

Attendees will learn about practices that should be avoided and followed in order to minimize potential biasing influences and will hear about the strategic, operational and political challenges of running a governmental crime laboratory independent of any law enforcement agency.

Examples from actual forensic casework in both criminal and civil cases will be used to illustrate the impact of bias on the outcome of forensic examinations and the manner in which such opinions are reported or expressed in court. Since *Daubert*, on remand to the 9th Circuit, implicitly recognized that cognitive bias on the part of experts in civil cases is a factor affecting admissibility, the means by which lawyers should seek to exclude proffered expert opinion that has been infected by the precursors of bias and to impeach expert testimony that may have been tainted by bias will also be discussed.

An experienced trial judge with extensive knowledge of forensic sciences will provide the gatekeeper's perspective, discussing how trial judges in both criminal and civil cases should deal with serious questions of cognitive bias on the part of forensic

experts. Gatekeeper decisions about admissibility, the use of cautionary jury instructions when serious issues of cognitive bias on the part of forensic experts have been raised during the trial, and whether to allow expert testimony from a qualified psychologist about the effect of cognitive bias on judgments made by forensic experts will be addressed.

Finally, a former state prosecutor who became a federal public defender will discuss the obstacles he faced as the criminal defense attorney representing Brandon Mayfield, the Oregon attorney wrongfully charged with the Madrid bombing, and the impact forensic bias had on him and his client.

Cognitive Bias, Forensic Experts, Judgment Errors

W11 Applications of Raman Spectroscopy for Trace Evidence Examinations

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Upon completion of this workshop, participants will have a better understanding of the principles of Raman spectroscopy, and a greater appreciation of the capabilities and applications of this technique for the characterization, comparison, and identification of the diverse materials that may be encountered as trace evidence.

This presentation will impact the forensic science community by providing knowledge, information, and hands-on experience of a theoretical and practical framework upon which participants can better utilize Raman spectroscopy for trace evidence examinations, and correctly interpret the spectral data thus obtained.

A number of significant developments in commercial dispersive and Fourier transform Raman spectrometers have occurred in recent years. They include holographic gratings, Charge-Coupled Device (CCD) detector arrays, efficient notch and edge filters to remove Rayleigh scattering lines, lasers covering a very wide range of excitation wavelengths from the ultraviolet to the near-infrared regions, and microscope attachments that allow spectral data to be collected from diffraction-limited spatial areas. Consequently, Raman spectroscopy is now a mature technique that is ripe for further applications in forensic science. In view of the ability of this technique to non-destructively probe very small areas with minimal sample preparation, Raman spectroscopy has considerable potential as another tool to characterize the diverse materials encountered as trace evidence. Such evidence may consist of complex matrices for which a battery of different analytical techniques is normally applied to obtain complete information about the properties of the various components. Despite this potential and the existing literature references demonstrating such applications, Raman spectroscopy remains an under-utilized technique in the forensic science laboratory. This workshop is intended to help rectify this situation.

The format of this workshop will consist of lectures in the morning and demonstrations and hands-on analyses in the afternoon. The lectures will cover various aspects of Raman spectroscopy. They include a brief theoretical background explaining the principles behind this technique, its advantages and limitations, and how the spectral data obtained by Raman spectroscopy can complement that produced by other techniques currently used for trace evidence examinations. In particular, the technique will be compared to infrared spectroscopy, as the two

methods both involve transitions of molecules to higher vibrational states, although this is affected by two very different processes. The Raman spectroscopic analysis of various types of materials commonly encountered as trace evidence, including fibers, paint, cosmetics, explosives, and general unknowns, will then be discussed.

For the practical exercises and demonstrations, several different commercial Raman spectrometers from different manufacturers will be available to illustrate the variety of instruments and accessories that forensic scientists can now choose from, the features of each, and the differences between them. Participants are encouraged to bring samples to the workshop for analyses.

Criminalistics, Raman Spectroscopy, Trace Evidence

W12 Root Cause Analysis — When Blaming the Analyst Completely Misses the Point

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After attending this presentation, attendees will: (1) understand corrective action and the basic construct of Root Cause Analysis (RCA); (2) learn the basic steps of performing an effective RCA; (3) learn the benefits and value, both monetary and organizationally, of performing a thorough RCA to determine what happened and why it happened; and, (4) learn why not to jump to conclusions and blame the individual.

This presentation will impact the forensic science community by providing the basic knowledge and skills to identify and determine the root cause of a problem to effectively implement appropriate corrective actions that will eliminate the problem. It is paramount that forensic science organizations take every opportunity to improve technical and management processes to ensure the quality of the forensic work product.

All organizations, regardless of size or mission, from hospitals, to toy manufacturing companies, to the aviation industry, are prone to problems, nonconforming work events, and departures from policies and procedures. Forensic science laboratories are no exception. However, the forensic science community must be ever so diligent in actively identifying, understanding, and correcting nonconforming events due to the impact that the quality of the work product has on the criminal justice community. Root cause analysis is a process used to define, evaluate, and systematically analyze a problem to determine the underlying reason(s) for the problem, the output of which is the input to corrective actions. Thus, it is essential for root cause analysis to be thorough for the corrective actions to be effective. If the analysis is incomplete or doesn't get to the "root" or underlying reasons, then chances are the problem will happen again. The philosophy of root cause analysis is in alignment with the philosophy of building a culture of quality and continual improvement. Every nonconforming event should be viewed as an opportunity for improvement of the management system. It is of the utmost importance for forensic laboratories to take every opportunity to improve technical and management processes to ensure the quality of the forensic laboratory work product.

During this workshop, the philosophy and purpose of root cause analysis will be defined and discussed. This presentation will outline the basic steps of root cause analysis and describe an effective approach for performing a root cause analysis. Examples

of ineffective and effective root cause analysis will be presented. Attendees will learn the process of asking “why” five times to get to the source of the non-conformance. In addition, participants will learn why “blaming the individual” is missing the point of the root cause process.

Forensic specific examples provided will include contamination in postmortem drug analysis cases after incomplete cleaning of a blender carafe and the Federal Bureau of Investigation (FBI) laboratory’s review of compositional bullet lead analyses cases. These examples will demonstrate how a thorough root cause analysis benefits the laboratory organization, the laboratory employees, and the laboratory customers.

Root cause analysis is a skill that must be learned, a process that requires continuous improvement, and a process that will require resources. It’s too costly, some might say. Are you willing to accept the risk of not doing root cause analysis well?

“A bad system will beat a good person every time.” ~W.

Edwards Deming

Root Cause Analysis, Continuous Improvement, Corrective Action

W13 Framework for Registration, Classification, and Evaluation of Errors in the Forensic DNA Typing Process

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The goals of this workshop are to encourage participants to accurately and truthfully record and document quality issues in their own forensic DNA laboratory and to teach attendees how to deal with such issues in the context of a case. A proper way to deal with errors is an essential tool to further improve on everyday forensic practice.

This presentation will impact the forensic science community by explaining how the precise magnitude of the error rate in forensic DNA typing is difficult to estimate, with the principal reason being the lack of a universally accepted definition of error in the professional society of forensic DNA typing laboratories.

Although DNA analysis is considered as one of the most reliable forensic tools available today, errors can be made during the course of the analysis. As this has a huge impact on the evidential value of a DNA match, there is a growing interest for actual data on the accuracy and error rates of forensic analyses and a more refined analysis of different types of errors and their causes.¹

In the report *Strengthening Forensic Science in the United States: A Path Forward*, the National Academy of Sciences refers to error rates as *misidentifications*: “proportions of cases in which the analysis led to a false conclusion (as the percent of incorrectly identified cases among all those analyzed).” The error rate includes both type 1 errors (wrongful reported match) and type 2 errors (wrongful reported exclusion). A major limitation of this approach is that the majority of errors in the DNA typing process do not lead to a misidentification. The consequence is that the majority of errors and near failures in the typing process will not be registered and will potentially stay undetected. The precise magnitude of the error rate in forensic DNA typing is therefore difficult to estimate, with the principal reason being the lack of a

universally accepted definition of error in the professional society of forensic DNA typing laboratories. The Netherlands Forensic Institute (NFI) has developed a comprehensive framework that allows for the classification, registration, and evaluation of errors in the forensic DNA typing process. In relation to the analysis of biological samples, the NFI has defined “internal quality issue notification” as *any* event that can lead to a failure or diminished quality of the analysis. These internal quality issue notifications have been benchmarked and evaluated using actual workload data from the department of Human Biological Traces of the NFI (over 400,000 DNA analyses) in the period 2008-2012.

This workshop will share data and the outcome of evaluations with the forensic community.

After attending this workshop, attendees will understand: (1) when an “internal quality issue notification” is made; (2) how an “internal quality issue notification” is made; (3) how “quality issue notifications” are assessed and evaluated; (4) how this can be used for benchmarking and process improvement; (5) how quality issue notifications are graded by potential impact and actual impact; (6) when and how the judicial system is informed; (7) when and how the public is informed; and, (8) how to deal with error rates in the context of a specific case.

In the first part of this workshop, an outline of the web-based NFI Quality On-Line Incident & Report Management system and an explanation of the procedures that allow for reporting quality issues in this system are given.² These presentations include details on the NFI work load, the data on the number of quality issue notifications over the years 2008-2012, and procedures on the assessment of quality issue notifications (necessary corrective actions taken, identification of the root cause of the quality issue, grading of notifications by potential impact, and actual impact). Also, essential benchmarking data on the performance of forensic DNA-typing in comparison with similar scientific disciplines (genetic testing centers) is presented.

The second part of the workshop focuses on impact analysis, explaining the framework that allows for an assessment and evaluation of the consequences of quality issue notifications for the conclusions of the DNA expert. Examples of errors with high and low potential and actual impact on the case will be presented.

The final session of this workshop discusses how the probability of an error affects the evidential value of a DNA match in a case. Discussion will include different views on how the DNA expert should incorporate the probability of an error in his or her report and will explain how the NFI deals with this.

References:

1. W.C. Thompson, “Tarnish on the ‘gold standard.’ Understanding recent problems in forensic DNA testing.” *The Champion*, 30(1): 10-16 (January 2006).
2. www.qualityonline.com

Error Rates, DNA, Laboratory Management

W14 Postmortem Monocular Indirect Ophthalmoscopy

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The goals of this presentation are to: (1) differentiate between direct and indirect ophthalmoscopy, noting advantages and limitations of each technique for the postmortem detection of fundal hemorrhages; (2) discuss the fundal location of retinal hemorrhages relative to their projected aerial image during monocular indirect ophthalmoscopy; and, (3) on a fundal diagram,

accurately draw retinal abnormalities observed during monocular indirect ophthalmoscopy with a simple ocular model.

This workshop will impact the forensic science community by providing an overview of postmortem monocular indirect ophthalmoscopy, facilitating skill acquisition, and evaluating practical training.

Postmortem examination of the retina has relied on ocular evisceration. In most medical examiner/coroner jurisdictions, ocular enucleation is not a standard autopsy procedure unless child abuse is suspected, thus creating observational bias when citing the prevalence of postmortem fundal findings such as retinal hemorrhages (preretinal, flame-shaped or splinter, and dot/blot) perimacular retinal folds, retinoschisis, and postmortem artifactual retinal folds. Postmortem Monocular Indirect Ophthalmoscopy (PMIO) permits examination of the decedent's posterior fundus and portions of the peripheral retina. The required equipment necessary for PMIO is relatively inexpensive and, when compared to direct ophthalmoscopy, the technique is less affected by corneal clouding, lens opacity, or vitreous hemorrhage and offers a wider field of view. PMIO uses a focal light source and an aspheric, convex condensing lens. An excellent source of coaxial illumination is a halogen or xenon surgical or procedural headlamp. This light source creates a collimated beam of light and permits the examiner to stabilize the condensing lens with both hands. Current aspheric lenses range from +14 to +40 diopters and come in different diameters, permitting a field of view of 35°-55°. Postmortem corneal opacity may cause the fundus to appear hazy; however, by gently removing the epithelial layer of the cornea, the emergent image is usually of adequate quality to readily detect lesions such as fundal hemorrhages and retinal folds.

Learning how to perform and become proficient at PMIO can be perplexing and intimidating. Most pathology residents and forensic pathology fellows have limited exposure to indirect ophthalmoscopy. Because the projected aerial image is inverted and laterally reversed, precise descriptions or recording of fundal abnormalities can be challenging. Unlike binocular indirect ophthalmoscopy with a teaching mirror attachment, an instructor and the fellow or resident cannot view the projected aerial image simultaneously during PMIO. To address these learning obstacles, it is necessary to develop tools and models to facilitate skill acquisition. An hour or two with an inexpensive ocular model can shift the learning curve of the resident, fellow, or forensic pathologist substantially to the right in how to correctly position the light source and hold the indirect lens.

This workshop consists of four sessions. An initial discussion and presentation reviews the technique of PMIO, highlighting the optics, the equipment, and examples of abnormal fundal findings found at autopsy by PMIO and the use of a smartphone to capture the projected aerial image. Next, attendees will have a realistic learning experience by practical hands-on training with a procedural headlamp, an aspheric indirect lens, and a simple ocular model containing a variety of retinal abnormalities observed at autopsy. The ocular models have variably sized "pupillary" openings and some will have clear acetate over the openings to simulate corneal glare. Attendees will be assisted in positioning the procedural headlamp, holding the indirect lens, viewing the projected aerial image, and accurately recording the retinal abnormalities. Attendees with smartphones can practice still image acquisition and video recording of fundal images produced by PMIO. Attendees will learn how to hold the smartphone with one hand while imaging the fundus and how to use a mini-tripod so the condensing lens can be stabilized with both hands, thus enhancing image stabilization and acquisition.

Following practice visualizing, diagramming, and image-capture techniques of numerous fundal images, attendees will finally be evaluated with a series of unknowns. Self-assessment of technical skill training and review of the unknown retinal findings

concludes the workshop. As part of the workshop fee, attendees will be given a USB thumb drive with the introductory presentation, sample retinal images, fundal diagrams, and articles on PMIO.

Forensic Science, Indirect Ophthalmoscopy, Retinal Hemorrhages

W15 Forensic Radiology: Current and Future Use in Medicolegal Death Investigation

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After attending this presentation, attendees will be able to discuss how current techniques in radiologic imaging are incorporated into medicolegal death investigations. In addition, they will be able to list the advantages and limitations of different imaging modalities as they relate to the potential to enhance a forensic autopsy. Workshop attendees will be able to outline issues which impact the expansion of forensic imaging into present practice models.

This presentation will impact the forensic science community by demonstrating how the advances in radiologic imaging are currently being integrated into medicolegal death investigations and offer potential for improving the efficiency and accuracy of forensic autopsy in the future.

Over the past century, basic radiologic imaging with film and fluoroscopy has been used in medicolegal death investigations. During this time, there have been numerous technological advances in the way radiographic images have been acquired and processed. As clinical medicine has validated their use in the living, pathology has been slow to incorporate them into the autopsy process. Currently, there are four radiologic imaging modalities that are used in medicolegal death investigations: fluoroscopy; radiography; Computed Tomography (CT); and Magnetic Resonance Imaging (MRI). The cross-sectional techniques, CT and MRI, are expensive and technically complex, which has been an obstacle to their use. However, they offer rapid visualization of anatomy that is difficult or nearly impossible to see during dissection. Additionally, they afford precise localization of foreign material of evidentiary value and facilitate its recovery. All modalities are currently available in digital format allowing for electronic recording, viewing, manipulation, archiving, and transmission of the images. While digital images offer timely viewing by all, they necessitate Picture Archiving and Communication Systems (PACS). Each modality has its own unique advantages and limitations with respect to medicolegal death investigations. A fifth modality, sonography, can be considered. There are limitations to this modality, mainly soft tissue air from either injury or decomposition that degrades the images; consequently, experience and incorporation into the medicolegal death investigation has been extremely limited.

This workshop will provide an overview of how the different radiologic imaging modalities have been incorporated into specific medicolegal death investigations. Through case presentations, the advantages and limitations of the modalities will be presented and how the images enhance the autopsy process will be discussed. Topics will include blunt force injuries, gunshot wounds, sharp force injuries, asphyxia, natural disease, fetal deaths, and postmortem changes and artifacts. Representative digital radiographs, CT images, and three-dimensional reconstructions will be presented for each topic area. Techniques that overcome some of the limitations of different modalities, such as postmortem angiography for the evaluation of vascular/soft tissue injury, the use of surface markers for the evaluation of gunshot wounds and sharp force injuries, and

the difficulty of MRI in ballistic imaging, will be discussed.

Personnel issues relative to advanced forensic imaging will be outlined. Who acquires images, interprets them, and how information is communicated to the forensic pathologist are issues of relevance to the present and future use of forensic imaging techniques. The final topic, possible future directions in the field of forensic radiology, will include discussion of possible training scenarios for forensic specialists, both physician and non-physician.

Forensic Radiology, Postmortem CT, Radiologic Imaging

W16 Science, Law, and the Inferential Process: The Epistemology of Scientific Conclusions and Their Role in the Legal Fact-Finding Process

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The goals of this presentation are to: (1) explore the epistemological structure supporting scientific conclusions; (2) illustrate how the inferential process engaged in during legal fact-finding may lead to conclusions contrary to those supported by scientific evidence; (3) illustrate methods utilized by science to measure the robustness of qualitative and quantitative results; (4) compare these scientific measures of confidence with legal burdens of proof and persuasion; (5) illustrate how scientific evidence can facilitate the determination of factual truth in the justice system; (6) demonstrate inferential tools meant to rationalize both forensic and legal determinations; and, (7) compare models of the inferential process in scientific and legal fact-finding contexts.

This presentation will impact the forensic science community by making explicit the inferential framework supporting scientific and legal determinations, revealing the distinctions and interplay between them. This will inform rule-making activities determining how and when scientific evidence can be utilized, help forensic and legal professionals better communicate scientific evidence to decision makers, and help scholars better understand how forensic evidence shapes legal determinations. This helps to maximize the benefits of forensic evidence and lead to more rational legal outcomes.

Both science and the criminal justice system strive to discover factual truth; however, in neither can we know what that truth is with complete certainty. Both systems are structured so as to provide information with specified characteristics. This information is relied upon as a basis for drawing necessary inferences. The inferences yield conclusions that may be believed to be correct but never known to be so. The process by which information is obtained and inferences are made determine the confidence we can have in those beliefs. The rigorous framework established by science permits the degree of certitude that can be accorded scientific conclusions to be quantitatively measured and expressed. The legal system provides less rigorous machinery which guides fact-finders in determining whether conclusions can be supported by designated burdens of proof. Even where scientific evidence is relied upon in the courtroom, the inferential processes engaged in are different in these two contexts. Because of this divergence, while forensic evidence often facilitates the determination of factual truth in the courtroom, fact-finders may rely upon it for conclusions

it does not support. This workshop will address the epistemological structure supporting legal and scientific conclusions and the interplay between the two when forensic evidence is relied upon.

The workshop will be broken down into two sections. The first session will examine the inferential framework supporting scientific knowledge/conclusions. Scientific knowledge/conclusions will be seen to be a matter of justified belief where models of our state of knowledge serve as incomplete and imperfect representations of physical states of interest. Measures of the epistemic robustness of these models and the conclusions they lead to will be illustrated in terms of degrees of belief expressing the uncertainty associated with a scientific result. Tools for, and models of, the inferential process in science will be developed and demonstrated and the meaning of scientific knowledge/conclusions will be explored.

The second session will examine fact-finding in the legal context when scientific evidence is relied upon. This process will be compared to that which occurs during the building of scientific knowledge. The manner in which scientific information is obtained by legal fact-finders will be examined along with the limitations and structural guides governing the analysis of such evidence. This session will consider architectural constraints of the criminal justice system where scientific evidence is concerned as well as modes and methods of practice. The nature of the conclusions reached by fact-finders and the degree of belief necessary to support different burdens of proof and persuasion will be compared to the measures of uncertainty relied upon by the sciences. It will be illustrated how distinctions between legal and scientific inferential processes make scientific evidence a resource that may either facilitate or potentially undermine the quest for factual truth in the courtroom. The workshop will conclude with a panel discussion addressing the day's subject matter with input and questions from attendees.

Disclaimer: The opinions or assertions contained herein are the private views of the authors and are not to be construed as official or as reflecting the views of the U.S. Department of the Army or U.S. Department of Defense.

Inference, Epistemology, Uncertainty

W17 Managing the 21st-Century Forensic Science Organizations

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After attending this presentation, attendees will be empowered to successfully resolve some of the most serious situations that arise in forensic management applications.

This presentation will impact the forensic science community by advancing forensic science policy and administration and improving the management priorities of our nation's forensic laboratories and offices.

The management of forensic science organizations in the 21st-century requires a unique blend of competencies and priorities. This workshop will assist attendees in developing specific strategies for better managing their organizations and setting the right priorities for managing time, costs, and efficiencies. Best practices, evaluative research, and contemporary management principles will be discussed with inclusion of case scenarios.

Forensic science administrators have a significant impact

on the criminal justice system. Therefore, they must function to the best of their abilities – all the time – and must be supported with quality information, sound recommendations, and access to enjoyable learning.

It is critical that agencies and managers overseeing a forensic science laboratory take their responsibilities seriously. At the top of any list for improving your forensic laboratory or office is effective communication. Making time to have constructive discussions with laboratory managers and staff about operations, policy and practice, laboratory efficiencies, and emerging opportunities and threats is of the utmost importance. Know what is going on in your laboratory and be prepared to make the difference.

Whether you are new to forensic science management or simply trying to steer your team in a new direction, optimism coupled with honest, self-critical analysis must be your focus. Know your people and their tendencies, gather as much information as you can, evaluate your options, and be decisive. Creating clarity about your role and purpose must be and must remain a critical priority for you and your management team. This workshop will help you set the right tone for your employees and customers.

The morning session will begin with topics that will help attendees produce organizational stability including: the meaning of high-impact leadership — what it means; business and leadership strategies; professional responsibilities; and influences of legal processes on your office (the Freedom of Information Act (FOIA), court orders, and subpoenas). The afternoon session will first focus on producing stakeholder commitment through customer service; accreditation and certification; competency and training; case management; and emerging best practices. Next, specific forensic manager perspectives with case examples will include a crime laboratory, a forensic toxicology laboratory, a quality-assurance manager, a coroner's office, and a medical examiner's office. The workshop will close with a panel discussion comprised of presenters and attendees.

Management, Efficiency, Best Practices

W18 Novel Psychoactive Substances (NPS): Pharmacology, Toxicology, Psychiatry, and Case Reports

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After attending this presentation, attendees will be able to: (1) identify several major drug classes within the group on novel psychoactive substances; (2) describe the pharmacological properties of the classes and the general pharmacological effects; and, (3) describe and categorize the adverse effect profiles and discuss the forensic significance of intoxication with these

compounds.

This presentation will impact the forensic science community by bringing together toxicological and behavioral sciences experts to provide complementary data on intoxications with novel psychoactive substances that are recently being implicated in assaults, homicides, suicides, and impairment-related traffic fatalities.

Over the past several years, a body of knowledge has begun to develop over Novel Psychoactive Substances (NPS) that are appearing on the market. Designer cannabinomimetic, stimulant, hallucinogenic, and depressant drugs comprise novel psychoactive substances. An overview of some of the resources available to researchers, laboratory professionals, forensic scientists, and care providers will be provided. Access to these resources help individuals gain further knowledge of the work being done on aspects of novel psychoactive substances outside their particular area of expertise, broadening their total understanding of the drugs and the implications of their increased use.

Since the recent increase in novel psychoactive substances, research has been focused on understanding the analysis, toxicology, pharmacology, or behavioral effects of the drugs. This workshop is intended to integrate the information on the toxicology and pharmacology of these compounds with knowledge that has been gained on the psychiatric implications as observed by psychiatrists, psychologists, and medical toxicologists. This will be achieved by pairing a discussion of the toxicology and pharmacology of various high-profile members of each of the drug classes with case reports in which there was analytically confirmed exposure. Information on the toxicology and pharmacology of individual drug classes such as tryptamines, cathinones, and synthetic cannabinoids will be covered to provide a backdrop for understanding the effects of these drugs and the legal implications of their use. Case reports on specific drugs such as methoxetamine, 5-MeO-DALT, and NBOMes will be presented. The adverse effects, forensic implications, assessment of patients, treatment, and outcomes will be considered.

This is a fast-moving and continually evolving area of drug use. The rapid pace of appearance of new compounds makes it essential for scientists to work together. By understanding the basics of the drug classes involved and specifics from case reports, individuals will be more prepared to quickly understand new compounds as they appear.

Novel Psychoactive Substances, Designer Drugs, Toxicology

W19 Serial Murder: Deconstructing Stereotypes

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After attending this presentation, attendees will have an understanding of serial murderers, their methods, motivations, and characteristics. The goal of this presentation is to expose attendees to recent comprehensive research on serial murder that has been conducted by the Federal Bureau of Investigation's (FBI's) National Center for the Analysis of Violent Crime (NCAVC), and the application of that research to actual serial murder cases, supplemented by serial murder case presentations and video clips of serial murderers discussing their crimes.

This presentation will impact the forensic science community by providing insight through the practical application of research into the dynamics and characteristics of serial murders and the offenders who commit these crimes.

Serial murder cases present numerous challenges and

obstacles to law enforcement personnel who have the responsibility of investigating these complex cases. These cases involve multiple victims; the series may span days, months, or even years; they can involve several jurisdictions; the motive involved may not be easily discerned; and there may be no obvious relationship between the offenders and the victims. Serial murder cases are also very rare and most law enforcement investigators do not have the same level of experience in investigating serial murders as they do with other types of crimes. Coupled with this rarity is the influence of the media and entertainment industries which have sensationalized serial murder and created exaggerated images of serial murderers.

The FBI's NCAVC is routinely consulted by federal, state, local, and international authorities in a variety of cases involving unusual, bizarre, or repetitive crimes, especially homicides. The NCAVC has traditionally been a front-runner in the study of serial murder. The original 1980's study of serial sexual murder conducted by NCAVC members was the catalyst for a plethora of research and study of serial murder by academicians, mental health practitioners, and law enforcement professionals. The NCAVC research and other subsequent studies have analyzed serial murders through a number of formats, including historical reviews, individual case studies, descriptive projects, and causality factors. Most of the studies were viewed in terms of causality, concentrating on the offender's development, upbringing, familial relationships, and history of physical, sexual, and emotional abuse.

While developmental information concerning a serial murderer provides interesting insights, it has little utility in helping identify an unknown offender during an active serial murder investigation. This is due to the wide range of factors involved in the upbringing and development of serial killers as well as the diversity among the serial murderer population. In an effort to bridge this gap, the NCAVC undertook a new serial murder research project. This project gathered information from solved case files to construct a database containing serial murder cases, the offenders who committed them, and their unfortunate victims. The results of that research project provides a perspective of serial murder that originates with how the victim's body was disposed, a known factor in most murder investigations. This allows investigators to identify a number of situational factors based upon the particular body disposal scenario, including the approach an offender used to gain access to a given victim, the motivation involved in the crime, and the nature of the relationship between the offender and the victim. Further, it helps to discern certain offender characteristics that assist in the identification of serial murder offenders.

This workshop will provide mental health practitioners, academicians, and law enforcement professionals with a basis for understanding serial murder based upon research and case examples from solved serial murder cases. It will also highlight the difficulties inherent in working serial murder cases and the need to understand the role of various law enforcement, laboratory personnel, and medicolegal professionals in these cases.

Serial Murder, Serial Murder Characteristics, Serial Murder Research

W20 Advances in the Investigation and Prosecution of Sexual Assaults Allegations

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After attending this presentation, attendees will be able to: (1) understand the role of the Sexual Assault Nurse Examiner (SANE) from medical examination to court testimony; (2) collect and preserve physical, psychological, and other evidence in sexual assault investigations; (3) become familiar with the Forensic Experiential Trauma Interview technique; and, (4) understand how collaboration among Sexual Assault Response Team (SART) members can impact the prosecution of the case.

This presentation will impact the forensic science community by informing attendees of the dynamics and proper procedures in the investigation and prosecution of sexual assault allegations and the necessity for a team approach for successful resolution.

Sexual assault and rape are crimes that occur in every socio-economic group and in every community around the world. Some of the disciplines charged with fighting this atrocity are the medical community, including: SANEs; the law enforcement community; the forensic laboratory community; and the judicial system. This workshop will examine each and discuss advances in these fields as they relate to sexual assault investigations and the prosecution of these cases. This workshop will emphasize each discipline on its own merits; more importantly, it will stress how every member of SART must work together to successfully resolve these allegations.

DNA plays a significant role in sexual assault investigations. Because of the intimate nature of sexual assault, biological evidence is often recovered during a sexual assault examination. SANEs should be aware of what types of items may contain useful DNA evidence, as well as where on the victim this type of evidence may be found, and how to properly collect and preserve it for testing.

Finding the suspect's DNA on a victim or at the crime scene can tie him to the scene, the victim and/or the crime. *Where* DNA is found and what type can also help to corroborate a victim's account of events. DNA testing has grown progressively more sensitive and robust over the past several years, enabling the successful typing of samples taken up to 96 hours post-event. Although a laboratory analyst will likely testify to the DNA results at trial, the SANE who collects it may also need to testify concerning how the samples were identified, collected, and packaged in order to satisfy chain of custody requirements.

Advances in SANE training and current best practices in evidence collection and preservation will be discussed. The importance of SANE collaboration with other SART professionals, especially the crime laboratory, and how it has played a significant role in holding the offender accountable will be presented. Additionally, this presentation will highlight how practices have changed and how evidence collection has impacted the progress of cases through the criminal justice system. Specifically, significant changes have occurred in the types of evidence collected, the manner in which it is collected, and the storage requirements of biological evidence as a result of advances in DNA identification, recovery, and analysis and the advantages of a team approach.

One of the many available and proven interview approaches is the Forensic Experiential Trauma Interview (FETI). This technique reduces the inaccuracy of the information provided, but also greatly enhances understanding of the interviewee's experience, increasing the likelihood of a better understanding of the totality of the event. The concept and approach of this technique can be described as a *forensic psychophysiological investigation*—an opportunity for the victim to describe the *experience* of the sexual assault, physically and emotionally. The FETI interview process obtains significantly more information about the experience, enhances a trauma victim's ability to recall, reduces the potential for false information, and allows the interviewee to recount the

experience in the manner in which the trauma was experienced.

Providing expert testimony is a part of any SANE's job. However, many medically trained professionals are uncomfortable in the courtroom. A familiarity with the legal process should help put them at ease and be better prepared to testify.

There are many legal issues related to sexual assault investigation and testimony. If a SANE and/or law enforcement officer collected evidence during the examination or scene search, they will need to testify to maintain the chain of custody. The SANE may have to testify to what the victim relayed during the examination if it is exculpatory, or it could be objected to as hearsay. Pre-trial preparation with the proffering attorney, as well as a thorough review of the case notes prior to testimony, should minimize courtroom nerves and maximize the testimony at trial.

Sexual Assault, DNA, SANE

W21 Effective Curricular Design and Improvement for Forensic Science Educational Programs

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After attending this presentation, attendees will: (1) be able to implement appropriate curricular design strategies for forensic science programs at the undergraduate and graduate level from defining appropriate and meaningful program goals and objectives, to linking these objectives to the curriculum, through to the use of effective delivery methods; (2) effectively use evaluation and survey research for program assessment and make meaningful program improvements from assessment measures; and, (3) design and adapt innovative curricular ideas based on those that have been successfully implemented in existing programs.

This presentation will impact the forensic science community by offering strategies for forensic science curriculum development and assessment which can be used to strengthen forensic science educational programs. This need has been acknowledged by practitioners and academics alike. This sentiment was mirrored in the National Academy of Sciences 2009 Report, *Strengthening Forensic Sciences in the United States: A Path Forward*, which stated that in order to "correct some of the existing deficiencies (in the field), it is crucially important to improve undergraduate and graduate forensic science programs."

Historically, forensic scientists came to the profession through traditional degree programs in various core scientific disciplines, typically in chemistry, physics, or biology. While these programs gave prospective forensic scientists a great grounding in the basic natural sciences, employers and educators felt that supplementing these core courses with forensic science principles would improve the quality of applicants to forensic laboratories. Instruction that provides an application of basic science to well-established forensic science disciplines could provide a broad view of the field of forensic science prior to specialization in a single area. This would hasten the time required for someone to be trained and

develop basic proficiency in a forensic science discipline once employed in the field.

Beginning around 2000, there was a proliferation of academic programs in the United States offering forensic science programs. They included everything from criminal justice degrees with a few crime scene and criminology courses, but very little science, to chemistry degrees with forensic science survey courses taught by non-practitioners. The profession recognized both a need for consistency in program content as well as an appropriate balance of basic and applied coursework emphasizing partnerships between academia and experienced practitioners. This led to the establishment by the American Academy of Forensic Sciences of the Forensic Science Education Program Accreditation Commission (FEPAC), which issued its first accreditation in 2004.

This workshop, sponsored by the Council of Forensic Science Educators (COFSE), a group of program directors and faculty from forensic science programs, is designed to help in the evolution of best practices for forensic science education, to identify creative teaching techniques and methods, to reinforce a research culture within forensic science education, and to promote the development of new degrees designed to meet the needs of forensic science agencies and the courts.

The program begins with considerations of curriculum design and introduces attendees to the concept of curriculum mapping, a process which allows schools to thoroughly examine and then revise their curriculum, encouraging programs to view their curriculum as a work-in-progress with the ultimate goal of providing the best preparation to students for careers in the field. This is followed by a consideration of the necessary components of bachelor's and master's degrees in forensic science, balancing and integrating graduate and undergraduate courses, and the optimum stage at which to introduce important forensic science concepts. The program includes several sessions on course design and delivery methods and encompasses key elements of a successful research project experience for both graduate and undergraduate students. How to engage and recruit practitioners to teach practical aspects within technical programs and to provide exposure to career choices for students is also featured in this part of the program.

In the second half of the program, the workshop will address tools and best practices for evaluating the student experience and assessing the effectiveness of the instruction, including the use of evaluations and survey instruments, followed by consideration of how to use that information in curriculum redesign and enhancement. Additional sessions will address opportunities to strengthen programs through study-abroad options, community engagement, and use of high-profile resources and programs such as a crime scene house. The program will end with a discussion of the pro's and con's of a professional doctorate in forensic science as opposed to the more traditional doctoral program approach that currently exists.

Education, Forensic Science, Curricular Design

W22 Questioned Document Examination and Enhancement of Evidence and Interpretation of Evidence Using Various Light and Filter Techniques

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After attending this presentation, attendees will have a better understanding of the theory and fundamentals in the examination of questioned document evidence using various light sources and optical filter image enhancement techniques. Attendees will also become aware of the various instrumental analyses, methods, enhancement techniques, and the interpretation of examination results.

This presentation will impact the forensic science community by equipping the attendees with the theory and practical knowledge in the areas of the examination and enhancement of questioned document evidence utilizing various light source and filter techniques.

Imaging of questioned document evidence has progressed from the use of the human eye to the analog camera, to the digital camera, to the use of the most sophisticated scientific instrumentation such as the use of Video Spectral Analysis (VSA) instruments. Whether it be from documenting faded handwriting to observing subtle ink differences, images available through a combination of modern-day filtering and application of various light sources can depict more about the appearance of evidence, especially when those image results come from image enhancement utilizing various types of light, filter, and wavelength variations to aid in enhancement and/or decipherment.

The modern-day questioned document examiner spends much of his or her time and effort making writing, typewriting, printing, alterations, and obliterations visible or more visible using various image enhancement techniques with the related images. These techniques for enhancement require fundamental and practical knowledge in the enhancement of document evidence utilizing various light sources and optical filter techniques, such as with the use of longpass, bandpass, long-wave and short-wave filtering methods, and their application, either individually or in combination in application of the examination of evidence. Upon completion of this workshop, attendees will have a better understanding of the concepts of utilizing these techniques.

This workshop will include a hands-on session following the introductory lecture on practical subject areas. The supplementary of practical exercises are intended to re-enforce hands-on participation of the attendees. Attendees will be provided with practical training in the fundamentals of the examination of questioned document evidence, using various light and filter enhancements techniques with the aid of instrumentation for the proper interpretation of examination results and limitations. The workshop will conclude with a discussion on the extent of questioned document evidence analysis of this nature and of enhancement techniques.

Questioned Document, Light Source, Optical Filter

W23 Applications of Next Generation Sequencing in Forensic DNA Analysis

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After attending this presentation, attendees will be able to understand next generation sequencing chemistries, principles, methodologies, and instrument platforms and their applications in forensic DNA analysis.

This presentation will impact the forensic science community by providing a forum for review and in-depth discussion on the utility of next generation sequencing technologies in forensic DNA analysis. Several new platforms have been developed for DNA sequencing that can be used for forensic DNA analysis. The attendees will also learn more about different applications of DNA sequencing technologies in investigating and resolving crime.

Short Tandem Repeat (STR) polymorphism and mitochondrial DNA sequence analysis are the most commonly used methods in forensic DNA laboratories worldwide. Fragment analysis, compared to DNA sequencing, has been the dominant method for determining STR length variation without sequence determination. This is mainly due to the ease of use and lower cost of fragment analysis compared to DNA sequencing. DNA sequencing is mainly used in mitochondrial DNA analysis to sequence hypervariable regions I/II of the mitochondrial genome. It is important to note that DNA sequencing has the potential to provide much more information compared to fragment analysis; e.g., sequence variation within STR region. DNA sequencing methodologies in use during the late 1990s were neither cost-effective nor simple enough to allow widespread investigation of Single-Nucleotide Polymorphisms (SNPs) and Insertions/Deletions (INDELs) for various identification and investigation applications.

Next generation DNA sequencing technology has transformed in the past decade. Multiple technologies are now developed that allow whole human genome sequence determination in a few days time compared to months. It is interesting to know that the next generation technologies are based on different biochemical principles such as monitoring released pyrophosphate upon incorporation of a base (pyrosequencing), fluorescence detection subsequent to reversible incorporation of a fluorescently labeled terminator base, a ligation-based approach wherein fluorescence of a cleaved nucleotide after ligation is measured, measuring the proton released after incorporation of a base (semiconductor-based sequencing), monitoring incorporation of a nucleotide by measuring the fluorescence of the fluorophore attached to the phosphate chain of the nucleotide, and by detecting the altered charge in a protein nanopore due to a released nucleotide by exonuclease cleavage of a DNA strand. The advancements in DNA sequencing technologies have resulted in reduced cost, ability to obtain sequence information from a few ng quantities of DNA, the ability to analyze multiple DNA fragments in parallel, availability of instrumentation from multiple commercial vendors, and relatively simple software for data analysis. These factors make adoption of next generation sequencing more attractive to forensic scientists and criminal laboratories than ever before.

Different applications developed using next generation sequencing include analysis for identity SNPs, lineage SNPs, Y-chromosome SNPs, phenotypic SNPs, mitochondrial DNA analysis, and STR analysis. The high capacity of next generation sequencing technologies enables simultaneous analysis of two or more types of genetic markers. For example, it is possible to obtain the results for mitochondrial DNA, autosomal STRs, Y-STRs, and SNPs simultaneously. Further, next generation sequencing brings a new avenue for mixture analysis and interpretation.

In this workshop, presentations will cover the following topics: (1) principles and chemistries of next generation sequencing technologies; (2) mitochondrial DNA analysis using next generation sequencing technologies; (3) SNPs and STR analysis using next generation sequencing technologies; and, (4) adaptation and validation of next generation sequencing technologies in forensic laboratories: the potential challenges.

For Research, Forensic, or Paternity Use Only.

Forensic DNA Analysis, DNA Sequencing, Next Generation Sequencing

W24 Applications of 2D and 3D Geometric Morphometrics in Forensic Comparisons

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The goals of this presentation are to: (1) provide an overview of Geometric Morphometric (GM) analysis; and, (2) demonstrate the methods for evaluation, measurement, and statistical comparison of shape change/distortion of an object in question. The generation of matching criteria and error rates based on a statistical model are discussed. Application of GM methods to bitemarks, fingerprints, and shoe prints to 3D visualization of 3D bullet comparison, medical forensics, and facial comparison will be presented. Advantages and disadvantages of the different approaches on comparison of shapes based on pattern recognition in digital images will be discussed.

This presentation will impact the forensic science community by showing methods that are used for pattern recognition in forensic comparison with error rates.

The goal of this workshop is to familiarize attendees with the possibilities for morphometric comparison in both 2D and 3D formats; the limitations and possibilities for use in forensic science are also discussed.

In 2012, the Forensic Sciences Foundation's (FSF) Theoretical Forensic Science Committee sponsored a contest, the "New Science or Technology to Forensic Science" competition. Based on the contest, this workshop was developed to discuss and present different aspects of morphometric comparison in forensic science, with application to odontology, footwear impressions, bullets, fingerprints, facial comparison, wound analysis, and 3D reconstruction. The workshop is updated with new solutions.

A well-developed method to describe shape variation between biological specimens is GM analysis. GM analysis involves placement of landmark points, curves, or outlines on either two- or three-dimensional images. The landmark data can be extracted and analyzed statistically as a unit. GM methods allow for a quantitative examination of shape by capturing the geometry of morphological structures of interest and preserving this information through statistical analysis. Shape information can be visualized by plotting landmark positions in Procrustes superimposition,

a method of optimally matching one shape to another. This can be performed with or without scale. Procrustes distances can be used to summarize variations in populations, to express the degree of similarity of individual specimens, means of populations, or to search for matches between specimens.

Among the tools available for statistical analysis is Principal Component Analysis (PCA) with which the principal variations of shape can be plotted and visualized. This allows for determination of which shape aspect is responsible for the most variation. Canonical Variates Analysis (CVA) can also be used to determine if shape information can distinguish between different categories of data. A range of other standard multivariate statistical methods can also be applied to shape data, allowing applications to a wide range of research questions and practical problems.

There has been much recent discussion in forensic forums concerning what constitutes a match or what defines two objects as being indistinguishable. It might be stated that two objects are identical when differences in measured attributes fall below measurement error levels, when the differences seen are no longer distinguishable from random effects. Error rates can be established by repeated measures studies in which the same object is measured multiple times. The Procrustes distance derived from GM analysis can be used as a quantitative descriptor of error and can be used as a threshold value below which objects might be said to match. Clearly, measurement resolution also depends on the scale of the object being measured and the resolution (smallest object measurable) of the means used to image the object. For example, in crime scene or accident reconstruction, the resolution required might be fractions of a meter; in a shoe print, individualizing detail might be separated by centimeters; in a bitemark, the achievable measurement resolution might be on a millimeter scale; fingerprints could be on a micron scale; and tool marks potentially might be submicron. With these examples in mind, careful attention to the issues of magnification (scale) and resolution is of the first importance; however, GM methods can be applied regardless of scale, as long as the nature of the data is understood. At each level of scale, error rates can be established and quantified. Using GM methods, large datasets can be statistically compared to explore the issues stated above. Ideally, quantification of the range and types of distortion produced will provide forensic practitioners with quantifiable validations of the quality of example items in the pattern evidence disciplines.

Morphometrics, Comparison, 3D



Criminalistics



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A1 Scanning Electron Microscopic (SEM) Analysis of Gunshot Residue (GSR) Collected From Human Hair Using a Vacuum and HEPA Filter

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After attending this presentation, attendees will understand the principles of Gunshot Residue (GSR) collection, the use of a Scanning Electron Microscope equipped with Energy Dispersive X-Ray (SEM/EDX) for GSR analysis, the potential for using a HEPA filter/handheld vacuum methodology to collect GSR from hair, and the feasibility of GSR sampling from hair as an additional method for screening suspects.

This presentation will impact the forensic science community by offering the possibility of augmenting the current method of GSR collection from suspects' hands with the use of a HEPA filter/vacuum methodology to collect GSR from hair. Identification of a practical approach for collecting GSR from hair could possibly offset the loss of GSR evidence from the hands due to daily activities.

This study began with the proposition that gunshot residue is present in hair, and that hair could be a useful source for collection of GSR evidence. However, before this hypothesis could be tested, a methodology for collection of particles had to be developed that resulted in effective capture of GSR. This experiment evaluates the usefulness of hair sampling in GSR collection as well as the effectiveness of the devised GSR collection methodology.

A series of experiments were conducted consisting of the collection of gunshot residue from three test subjects' hair and hands after firing a different caliber handgun. One handgun used total copper jacket bullets, one used jacketed hollow-point bullets, and the third used lead round-nose bullets. The collection method included GSR stubs and, for the hair collection, the addition of a handheld vacuum and a HEPA filter. The GSR stubs were analyzed using SEM/EDX.

The handheld vacuum was chosen because it was lightweight, rechargeable, portable and most of all, highly efficient in collecting dispersed microscopic particles. This simple approach of using a handheld vacuum with a HEPA filter attached to it has the potential for future use in crime scene investigation kits. It also proved feasible to recover GSR from the filter using an adhesive disc that was analyzed by SEM/EDX.

The lead round-nose bullets produced the most significant amount of GSR both in the hair and on the hands, with the jacketed hollow-point bullets producing the second-most abundant amount of GSR, and the total copper jacketed bullets producing the least. The GSR on the hair of the lead round nose bullets was detected throughout the entire experiment including the final three hour mark; however, common elements usually associated with GSR were the only elements found on the hands at the three hour mark. Lead, barium, and antimony (Pb-Ba-Sb) is such a unique particle that when found, it can directly be connected to GSR. Other characteristic compositions including Pb-Sb, Pb-Ba, Sb-Ba, Sb, Ba,

and Ba-calcium (Ca)-silicon (Si) (with trace sulfur) are not unique but are suggestive of GSR. The collected results suggest that daily activity removes the particles and makes it harder to detect in the hours that follow a firearm discharge, and that hair sampling method is a viable possibility as an additional method of detection.

The results indicate that it is feasible to detect gunshot residue in human hair using a HEPA filter and a vacuum. The HEPA filter allows the screening of particles as small as 1-10µm. The filters used in this experiment screened particles as small as 0.3µm. Because portability is a necessary requirement for crime scene equipment, the vacuum used was a rechargeable handheld model, and it proved to have enough power to gather GSR particles from hair. These results show that SEM with a highly sensitive energy-dispersive X-Ray detector enables the sampling of both human hair and hands to yield more conclusive forensic findings than hand swab evidence alone.

Gunshot Primer Residue, Hair, SEM/EDX

A2 The November 2, 2007 Murder in Perugia: One Thousand Clues of Evidence That Tell Another Truth — What About Justice for Meredith Kercher, Raffaele Sollecito, and Amanda Knox?

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After attending this presentation, attendees will understand the relevant role of scientific evidence in the trial and how evidence was evaluated in the homicide investigation of Meredith Kercher.

This presentation will impact the forensic community by providing an example of how several strands of evidence were used in the reconstruction of the assault perpetrated on the victim.

The main goal of this presentation is to comprehensively characterize the relevant forensic features of the case and their importance for case reconstruction and judgement in terms of Italian and international laws.

The interpretation and evaluation of classical scientific proofs in a trial are set at a considerably higher level of value than the simple correlation of clues. Actually, these often fail to achieve a sufficient level of certainty in a judgment. Moreover, the evaluation systems of a judge and a scientist are obviously very different. The court must, in fact, express its opinion even when the scientific evidence is insufficient, and thus judgment, unfortunately, is sometimes wrong because it is based mostly on a deep conviction of guilt.

Professor F Vinci was Sollecito's forensic expert from the onset of the trial for the murder of Meredith Kercher. He followed the case over its course in which Amanda Knox, one of Meredith's roommates, was also involved. In this presentation, different and controversial pieces of evidence will be used to illustrate and

reconstruct a sequence of events based on shoe and footprints, the luminol investigation, bloodstains, examination of a pillow, knife identification, and so forth.

Many investigations were carried out by several experts in various scientific fields, who served as consultants to the prosecutor and the defendants' attorneys. Unfortunately, the results, often discordant, were debated in the trial.

Some problems of international law and agreements on extradition protocols between the U.S. and Italy will also be discussed, as well as some fundamental legal principles on the validity of evidence in criminal trials in Italy.

This presentation will make a positive contribution to the forensic community by presenting a case study in which the analysis of injuries along with their anatomical location, the results of crime scene investigations, and the results of laboratory investigations were insufficient to eliminate the many doubts that still surround the murder of Meredith Kercher in Perugia. This uncertainty has resulted in the initial judgment being overturned on appeal. A second trial is due to be heard in the Italian Supreme Court.

Meredith Kercher Homicide, Stab Wounds, Forensic Pathology

A3 Degradation Parameters of the Aging Process of Latent Fingermarks

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After attending this presentation, attendees will have an inside view of the latest research being conducted on latent fingermarks degradation processes based on observational physical indicators.

This presentation will impact the forensic science community by attempting to determine, as precisely as possible, the time that evidence of a biological origin was deposited at a crime scene.

Forensic science is a multidisciplinary field that utilizes scientific principles and techniques for the analysis of physical evidence at crime scenes and the subsequent identification of donors of origin. Its main goal is to obtain valid evidence with maximum objectivity and reliability to provide to the justice system. Technical and methodological developments in the identification of latent fingermarks have been at the center of forensic science since its inception in the 19th-century. Over time, these advances have included an extensive range of development reagents for use in many types of environmental and substrate conditions and tools such as automated fingerprint identification systems. However, issues involving the reliability of latent fingermarks frequently arise in court proceedings during presentation as evidence when they

cannot be directly correlated to the moment a crime was committed. A methodology that determines, as precisely as possible, the time that evidence of a biological origin was deposited at a crime scene would be of great value to law enforcement agencies and penal courts; this would provide them with the ability to more accurately place a suspect within the time frame of an alleged crime beyond any subjective witness or victim statements. Intensive research is being conducted around the world by government, academic, and private organizations to determine, through chemical means, the precise time of latent fingermark deposition on different types of surfaces. Unfortunately, these approaches usually involve expensive equipment and complex techniques that are time consuming and require great expertise by highly skilled researchers.

The present proof-of-concept study introduces an inexpensive quantitative method that attempts to empirically establish the aging process of latent fingermarks based on observational physical indicators, taking into account the wide range of influencing factors involved. This approach consists of obtaining, as precisely as possible, the deposition time of fingermarks visualized by the application of common physical developers used in the field, which are easy to apply, inexpensive, and do not require sophisticated forensic techniques or a scientific expert. The study aims to reveal particular visual patterns of degradation of latent fingermarks after being exposed to certain monitored indoor laboratory environmental conditions. Factors considered include temperature, relative humidity, air currents, type of fingerprint depositions (sebaceous and eccrine), various exposures to daylight (direct, penumbra, and darkness), and type of physical substrate (glass and plastic) over a continuous aging period of six months in 11 discrete development times. Statistical data has been obtained from the quantifiable analysis of four visual parameters commonly observed in the progression of a fingermark aging process: reducing width of ridges, decreasing color contrast between ridges and furrows, increasing number of discontinuities, and decreasing minutiae count. Furthermore, the study also aims to identify any potential correlations between environmental conditions and measurable parameters. Preliminary data, still under development, allow investigators to discriminate the distinct aging processes of fingermarks exposed to different environmental conditions, each with unique evolution patterns. Early results in the analysis suggest the method can be used to narrow the time frame of unknown fingermarks' depositions to within three months. Once the study is complete, it is expected to reduce this window to a few weeks or to several days.

As a procedure that is economically affordable to most agencies, the described scientific method would strengthen the probative value of criminal evidence while saving costs in the laboratory and reduce the amount of additional criminal proof required during judicial processes.

Degradation, Parameters, Fingermarks

A4 Effects of Different Heat Exposure Time and Temperature Profiles on the Chromatographic Profile Derived From Porcine Samples Under Laboratory Conditions

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After attending this presentation, attendees will: (1) gain a better understanding on the process of generating thermal

decomposition products from porcine samples under laboratory conditions; (2) be familiar with the type of products generated from the thermal degradation of porcine samples; (3) identify the effects of the length of exposure to heat on the chromatographic output and nature of the chemical products generated; and, (4) identify potential differences in chromatographic data generated from fleshed and defleshed porcine tissues under the above-mentioned conditions.

This presentation will impact the forensic science community by illustrating how to identify the differences in the nature and identity of compounds generated from porcine samples at different temperatures and at different durations of heat exposure under laboratory conditions. The outcome of this study will contribute to the fire investigation community through the provision of an increased understanding of the laboratory analysis of cadaveric samples in fire-related cases by providing a better perspective on the effects of temperature and duration of exposure to heat on such samples. Information from this study could potentially aid in the interpretation of fire debris data derived from scenes involving human remains.

While studies have shown that certain temperatures affect the detection of volatile products from porcine samples, this study is based on previous work by systematically monitoring the derivation of such products under controlled laboratory conditions.¹ Fleshed and defleshed porcine samples were heated to the point of ignition and then allowed to burn for different durations. The temperature and length of exposure to different temperatures were recorded for each sample type. The samples were extracted using passive headspace analysis whereby the derived volatile products were adsorbed onto Activated Carbon Strips (ACS). The strips were then desorbed in solvent and analyzed using Gas Chromatography/Mass Spectrometry (GC/MS) according to ASTM guidelines.

Significant differences were observed in the chemical profile of the target samples as the temperature to which the samples were exposed and the duration of that exposure were adjusted. Variation in temperature and exposure durations were evaluated, based on the type and consistency of chemical products generated at each sampling stage across a set of six repetitive samples for each test. Results for both samples were relatively similar and indicated different sets of volatile products attained at temperatures before auto-ignition (<250°C) and at temperatures beyond auto-ignition (>300°C). The former predominantly generated a homologous series of n-aldehydes, whereas the latter produced a variety of n-alkanes, n-alkenes, and aromatics.

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Pyrolysis, Temperature, Heat Exposure

A5 Evaluation of the Degradation of Lotion Components Due to Age and Exposure

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After attending this presentation, attendees will gain a broad understanding of the forensically significant chemicals which comprise commercial lotions and how the compositions of the lotions can vary due to age and exposure.

This presentation will impact the forensic science community by providing the knowledge base to examine and compare commercial lotions with residues collected from crime scenes, clothing, and other evidence. An evaluation of the

variability between commercial lotions was explored, as well as the effects of age and exposure on the chemical constituents of lotions, thus providing attendees with the knowledge to effectively interpret analytical results.

Commercial lotions are ubiquitous in modern society. Lotions and their residues may be encountered in forensic casework in a variety of fashions; by the transfer to items of evidence through contact in assaults, theft, breaking and entering, and sex crimes. Evaluation and comparison of lotion products in the possession of the suspect or victim with residues from evidence can provide a powerful evidentiary link.

Commercial lotions consist of complicated mixtures of materials both synthetic and naturally derived. Lotion formulations contain a wide variety of materials which are present for various end uses. Most lotions contain a lubricant or vehicle that acts as a delivery system for the other ingredients. Ingredients are added to function as emollients, acting to soothe and soften the skin. Fragrance compounds may be employed. Various UV-absorbing compounds may be found which are used to protect the skin and to prevent the degradation of the other lotion ingredients. Finally, lotions may employ materials for other aspects of skin care, such as Vitamin E or aloe. In addition to the materials that are found as listed ingredients, additional chemical compounds were detected which may originate from contaminants in the source materials, or which may be formed during the manufacturing process. Because of the large number of different materials that may be encountered in a lotion sample, a high degree of specificity may be obtained from comparisons of these samples.

In this study, a sample set of over 100 commercial lotion products was analyzed using Polarized Light Microscopy (PLM), Fourier Transform Infrared Spectroscopy (FTIS), Gas Chromatography/Mass Spectrometry (GC/MS), and Direct Analysis in Real Time-Mass Spectrometry (DART®-MS) to assess the degree of variation between different products. The results of these analyses can provide numerous data points from which a comparison to a known source or questioned sample may be made. The largest of variability was found in the organic composition of lotions and was observed using mass spectrometry techniques. This data provided the most discrimination between samples. Some samples differed by the general class of material included in the composition (parabens, alcohols, fatty acids, etc.), while other samples differed by the specific components within each class (e.g., ethylparaben vs. propylparaben).

Factors such as the presence/absence of certain chemical constituents, relative concentrations of various components, and frequency of occurrence of the chemical components were evaluated. The frequency of occurrence was evaluated to determine the most discriminating components in lotion samples, and principal component analysis was performed to assess the discriminatory value of the components detected in lotion samples.

Inter- and intra-lot variability was also assessed. Several samples of the same product line, but of different lots, were collected and analyzed from several different manufacturers. Duplicates of each lot sample were also analyzed. Some minor variation was observed between different lots of the same product, which may be due to variations in raw materials or manufacturing location. A full discussion will be provided.

Finally, the effects of aging and exposure were evaluated on the lotion samples from the inter-/intra-lot variability examination. Samples of the lotions were allowed to age with exposure to atmospheric conditions, and samplings were retested after regular intervals. Noticeable changes in the composition were detected, particularly in the presence and concentration of volatile compounds (e.g., propylene glycol, phenoxyethanol, and fragrances) as well as in the concentration of UV-absorbing compounds (e.g., parabens). An evaluation of the constituents which were lost due to evaporation and exposure will be discussed,

providing insight that can be utilized for data interpretation in the performance of forensic science casework. A casework example will be highlighted to demonstrate the specificity obtainable using this analytical scheme.

Commercial Lotions, Mass Spectrometry, Trace Evidence

A6 Optimization of Development of Latent Fingerprints on Unfired and Fired Brass Cartridge Casings

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After attending this presentation, attendees will understand the optimal sequence of latent fingerprint development techniques to use on unfired and fired brass cartridge casings. Before a bullet and its casing are loaded into a gun, it is presumably handled and marked with fingerprints. After the bullet has been fired, its casing is ejected from the gun. If not retrieved, this casing is left at the scene. Cartridge casings found at crime scenes may contain fingerprint evidence to introduce or help convict a suspect; however, there are few articles on the subject of the best method to develop latent fingerprints on cartridge case evidence.

This presentation will impact the forensic science community by providing knowledge of useful processing techniques as this research compared the best development methods for unfired casings found in previous research, along with other commonly used development methods.

After the most effective method was discovered, that method was applied to fired brass cartridge casings to determine the likelihood of developing a clear fingerprint. For Phase I of this research, five different methods and 500 unfired .40 caliber brass cartridge casings with one sebaceous fingerprint on each casing were used. The methods used were: cyanoacrylate fuming followed by BY40 followed by magnetic black powder, cyanoacrylate fuming followed by gun blue followed by BY40, cyanoacrylate fuming followed by magnetic black powder, cyanoacrylate fuming followed by BY40 followed by acidified hydrogen peroxide, and cyanoacrylate fuming followed by magnetic black powder followed by acidified hydrogen peroxide followed by BY40. The two techniques that yielded the best results were cyanoacrylate fuming followed by gun blue followed by BY40 and cyanoacrylate fuming followed by BY40 followed by acidified hydrogen peroxide. The technique that produced the highest mean number of clear fingerprints was cyanoacrylate fuming followed by gun blue followed by BY40; this method was chosen to be used in Phase II of the research and applied to fired cartridge casings. Phase II consisted of firing 50 .40 caliber bullets from a .40 caliber handgun after one sebaceous fingerprint was placed on each casing. Although cyanoacrylate fuming followed by gun blue followed by BY40 yielded clear fingerprints with unfired casings, this method did not produce any clear fingerprints on fired cartridge casings due to the changes a casing undergoes during the firing process.

Previous research has emphasized using other effective development methods but consist of techniques not easily applied by the majority of the forensic science organizations. It is important to discover successful development methods that are practical and accessible. Useful methods that can be used to develop latent fingerprints on brass cartridge casings in any forensic laboratory will be presented.

Fingerprints, Casings, Development

A7 Fluorescence From Latent Fingerprints Irradiated by Visible Light

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After attending the presentation, attendees will gain a new understanding of what is fluorescing when latent fingerprints are irradiated by visible light.

This presentation will impact the forensic science community by demonstrating that a portable Continuous Wave (CW) green laser can effectively visualize latent fingerprints and by suggesting that heating may become a new pretreatment technique in crime scenes.

It is believed that fingerprints stem from secretions released from eccrine, apocrine, and sebaceous glands.¹ In order to measure fluorescence from the secretions, the optimum excitation wavelength is considered to be the ultraviolet wavelength shorter than 380 nm.² It has been reported that keratine, one of the major proteins included in nail and epithelial cells, fluoresces by irradiation with visible light and the fluorescence intensity increases when heated.³ Advanced Glycation End products (AGEs) in the skin, which are produced via glycation of protein (i.e., non-enzymatic combination of protein with reducing glucose), have fluorescent properties similar to keratin.⁴ All of them have not been fully utilized for visualization of latent fingerprints. In this study, the hypothesis that latent fingerprints have at least some of these as residues and, therefore, fluoresce by irradiation of visible light was tested.

The following samples were prepared: a piece of human skin, a filter paper with an impression of the skin piece, and another filter paper with an impression of fingerprints. A 532nm CW laser was used as an excitation source in the visible wavelength region. Its output power was set to be 1.98W. Fluorescence was measured by a hyperspectral imager with an attached 560nm long pass filter to reject the excitation laser.

Averaged fluorescence spectrum of the skin piece had its maximum at 620nm. Therefore, if necessary, the single-wavelength image was constructed using fluorescence intensity averaged over the band centered at 620nm with a band width of 10nm. The maximum intensity of averaged fluorescence spectrum of the heated skin piece increased as expected. The single-wavelength image of the filter paper with the skin piece impression demonstrated that the impression itself fluoresced. The image of the fingerprint impression on the filter paper was clearly visible. The present results demonstrated that latent fingerprints could be visualized by the 532nm CW laser.

Advanced glycation of protein increases fluorescence intensity. Heating accelerates glycation of protein.³⁻⁵ If proteins glycosylated at various levels are included in latent fingerprints, it is suggested that heating may be regarded as a new pretreatment technique in order to visualize latent fingerprints by irradiation with visible laser.

In conclusion, the present results demonstrate that fingerprints indeed fluoresce under visible laser irradiation and suggest that heating may become a new pretreatment technique for the enhancement of latent fingerprints.

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Fluorescence, Advanced Glycation End Product, Green Laser

A8 TLC-SERS for the Identification of Controlled Substances

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After attending this presentation, attendees will understand the potential of Thin Layer Chromatography with Surface-Enhanced Raman Spectroscopy (TLC-SERS) for the identification of controlled substances.

This presentation will impact the forensic science community by providing a rapid, reliable, and sensitive technique for identifying illicit drugs through the use of TLC-SERS.

The combined method of Thin Layer Chromatography and Surface-Enhanced Raman Spectroscopy (TLC-SERS) is ideal for the separation and identification of controlled substances and controlled substance mixtures. TLC-SERS is an ideal technique because it requires less analysis time and smaller sample sizes when compared to other methods of drug analysis. In addition, it adheres to the current standards set forth by the Scientific Working Group for the Analysis of Seized Drugs (SWGDRUG). This method for separating and identifying illicit drugs involves separating mixtures on a TLC plate then, through the addition of a metallic colloid, identifying the components directly from that TLC plate using Raman spectroscopy.

TLC is a separation process that is commonly used as a screening test in forensic laboratories because it is rapid, inexpensive, and efficient when separating components of a mixture.

Raman spectroscopy is an identification method that examines the frequency change of a laser light source due to its interaction with a sample. Inelastic scattering occurs when the frequency of photons in the laser change after the photons interact with a molecule, and this vibrational frequency depends on the molecule's structure. The advantages for this identification technique are minimal sample preparation, accurate molecular identification, and non-destructive analysis. The major limitations to Raman spectroscopy are low sensitivity and fluorescence interference.

SERS essentially corrects for the disadvantages of normal Raman spectroscopy. SERS mirrors the procedure of Raman — the main difference being the addition of a silver nanoparticle colloid directly onto the sample, which, in this research, are the separated spots on the TLC plate. The addition of a metallic colloid enhances the scattering signal of the molecules and allows for spectra to

be collected from samples that cannot be detected using normal Raman spectroscopy. A SERS spectrum can be acquired quickly and includes all the advantages of normal Raman spectroscopy. SERS spectra are unique to single compounds and therefore allow this method to be used as a technique for identification.

In this research, TLC-SERS was shown to be successful for the separation and identification of illicit drug mixtures containing cocaine, methamphetamine, MDMA, and codeine. Because TLC is already completed as a screening tool, the combination of SERS for identification directly on the TLC plate would not require the use of additional sample and only minimal additional sample preparation and analysis than is currently employed for the forensic analysis of controlled substances.

Controlled Substances, TLC-SERS, Raman Spectroscopy

A9 Direct Identification of Explosives Residues on Banknotes Recovered From an ATM Explosion Using Easy Ambient Sonic Spray Ionization Mass Spectrometry (EASI-MS)

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After attending this presentation, attendees will understand how Easy Ambient Sonic Spray Ionization Mass Spectrometry (EASI-MS) can be applied to the analysis and identification of explosive traces on banknotes, helping to solve crimes involving Automated Teller Machine (ATM) thefts.

This presentation will impact the forensic science community by teaching how EASI-MS can be used to significantly reduce time spent in analysis and identification of traces of explosives on banknotes and increase the specificity of the screening process.

ATM thefts, particularly involving the use of explosives, is a new type of crime in Brazil and is increasing. According to Police Intelligence Department from São Paulo in Brazil (DIPOL) data, the estimate is that 500 ATMs were stolen in 2011 just in São Paulo State. On average, each ATM contains between R\$70,000 and R\$100,000. If the thieves were successful in taking the money from all 500 stolen machines last year, the gangs have acquired approximately R\$50 million. Thus, the analysis and identification of traces of explosives on banknotes are very important because they provide scientific and technical elements to frame the notes as a result of bank robbery-constituted evidence for police investigations and the proper classification for the law and the application of justice. In this work, EASI-MS is a fast ambient ionization source that could significantly reduce time spent in analysis and identification of explosive traces on banknotes and increase the specificity of the screening process.

EASI-MS data in the negative and positive ion mode was collected at the ThoMSon Mass Spectrometry Laboratory using either a mono-quadrupole mass spectrometer or a Fourier Transform Ion Cyclotron Resonance Mass Spectrometer (FT-ICR/MS).

Preliminary data results indicate that explosives samples of trinitrotoluene (TNT) and picric acid (TNP) were observed as

(M-H) anion and trinitrohexahydro-1,3,5-triazine (RDX), octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine (HMX), pentaerythritol tetranitrate (PETN), and nitroglycerin (NG) were observed in the negative ion mode as chloride adducts detected and identified in a few seconds on the different standard surfaces (glass, metal, and paper) with no sample preparation, by EASI-MS using a spray solution of methanol–water. Real banknote samples collected directly from an ATM explosion in São Paulo-Brazil were investigated in the negative and positive ion mode in order to identify such explosives and their decomposition products.

EASI-MS, Explosives, Banknotes

A10 Optimized Extraction and Separation of Synthetic Cannabinoids in Herbal Incenses

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After attending this presentation, attendees will learn the optimized extraction methods for the study of synthetic cannabinoids in herbal products as well as High-Performance Liquid Chromatography (HPLC) separation and quantification with Diode Array Detection (DAD). Attendees will also be informed about the principles of Reversed-Phase Liquid Chromatography (RPLC) with UV-Vis detection as it pertains to forensic chemistry and the optimized procedure for extracting and identifying synthetic cannabinoids in herbal incenses. Through the comparison of various solvents, agitation methods, and filtration techniques, the optimal extraction protocol for synthetic cannabinoids can be determined and adopted in a practical setting.

This presentation will impact the forensic science community by providing an optimized and low-cost sample extraction method and HPLC quantification methods for the accurate and speedy determination of synthetic cannabinoids in herbal incenses.

In addition to a dozen synthetic cannabinoids, recently three new cannabinoids were added to Schedule I controlled substance list under the Controlled Substance Act. It remains a great challenge for forensic scientists to efficiently separate, quantify, and identify these “cannabimimetic agents” in herbal incenses due to the similarity of isomers or analog structures. Newer compounds are being synthesized promptly to circumvent the ban, which exacerbates the analytical difficulties for forensic labs, some of which are still experiencing backlogs. This demonstration will provide the forensic science community a simple, quick, and low-cost technique for the extraction of synthetic cannabinoids in herbal incense in order to improve the accuracy and throughput of “Spice” evidence processing.

A mock “Spice” sample was created in a manner that imitates its production in clandestine laboratories. An extraction protocol was optimized to yield the highest efficiency with the lowest cost. Various solvents, mechanical agitation methods, and filtration techniques were compared and contrasted. Following the extraction, HPLC separations with DAD were performed to separate and quantify the synthetic cannabinoids in the herbal extracts. The separations were conducted on an Agilent® 1100 HPLC with phenyl functionalized columns, conventional C-8 and C-18 columns.

After the separations, the best combination of extraction variables was determined to be methanol as a solvent, along with manual agitation to free the cannabinoids from herbal base, and paper filtration. The method consistently extracted approximately 40% of the cannabinoids from the herb. The retention times were

used to identify cannabinoids in the herbal sample and peak areas were used for quantification through external calibration. It was found that cannabinoids including XLR-11, AM-2201, UR-144, JWH-122, RCS-4, and RCS-8 were regularly added to herbal incenses with concentrations ranging from 0.5mg to 30mg per gram of herb. The powder was not sprayed evenly as the relative standard deviation from multiple batches often exceeds 20%. The identity of these cannabinoids was confirmed with both UV-Vis spectra and Gas Chromatography/Mass Spectrometry (GC/MS). Compared to the phenyl functionalized columns, a C-18 column was found to be just as effective at separating the cannabinoids.

In conclusion, simple, low-cost methods can be used to yield a high synthetic cannabinoid extraction yields from herbal incense. This method, when paired with RPLC and DAD detection can be used to quickly separate, identify, and quantify synthetic cannabinoids in “Spice” samples in the laboratory.

Synthetic Cannabinoids, Extraction, HPLC

A11 Degradation of Synthetic Cannabinoids UR-144, XLR-11, PB-22, and 5F-PB-22 Analyzed by UPLC-QTOF Mass Spectrometry

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After attending this presentation, attendees will be aware of potential degradation products from synthetic cannabinoid compounds UR-144, XLR-11, PB-22, and 5F-PB-22. This presentation introduces products that could potentially be formed prior to the ingestion of the intended compounds, along with a method utilizing Ultra-Performance Liquid Chromatography Quadrupole Time-of-Flight Mass Spectrometry (UPLC-QTOF/MS) for the identification of degradation products.

This presentation will impact the forensic science community by providing information related to the structural stability of some synthetic cannabinoids and notifying practitioners of related degradants that users or crime laboratories may encounter.

Synthetic cannabinoids can be acquired through multiple routes, including purchase via the internet. They are typically ingested by either vaporization of the raw compound or smoking a cigarette composed of the cannabinoid dissolved in solvent, then sprayed on plant material. Compounds UR-144, XLR-11, PB-22, and 5F-PB-22 have all been found on the market with little to no information available on their stability.

In order to identify degradation products to which users may potentially be exposed, the synthetic cannabinoids were subjected to forced degradation studies. Samples at approximately 1mg/mL concentration were prepared in acidic, basic, and oxidizing solutions, and placed in a stability chamber at 25°C, 65% relative humidity. Around 1mg of each cannabinoid was placed in an individual clean vial and stored in a 105°C oven for temperature-induced degradation. Samples were stored in their respective storage conditions for 0, 24, 48, 72 or 96 hours, at which times aliquots were taken out, diluted in methanol, and stored at -80°C until thawed and analyzed.

Data were collected using a Waters Acquity UPLC interfaced serially to both a Photo Diode Array (PDA) detector and a Waters Synapt G2 QTOF/MS operated in positive ion electrospray mode. Liquid chromatography was carried out using an Acquity BEH C18 column (1.7µm X 2.1 X 50mm). A gradient elution method with a flow rate of 400µL/min was used with mobile phase

A consisting of water with 0.1% formic acid and mobile phase B consisting of methanol with 0.1% formic acid. The amount of degradation achieved under each condition was determined based on comparison of the analyte LC-PDA peak area to that of a freshly prepared control sample made from the same test batch.

Significant degradation was observed in both UR-144 and XLR-11 samples stored at elevated temperature. After 96 hours at 105°C, at least 50% of the original UR-144 analyte was converted to a single degradant with a retention time 0.2min earlier than UR-144. MS data indicate that the degradant has the same molecular weight as UR-144 and a similar fragmentation pattern. This degradant was determined to be an isomer of UR-144 in which the cyclopropyl ring is opened, based on retention time and fragmentation match to a reference standard. Similar results were seen for XLR-11. These degradants are likely to be encountered in raw material that has been exposed to high temperatures during shipping or storage, or formed in the GC/MS instruments commonly used for analysis in forensic laboratories.

PB-22 and 5F-PB-22 proved resistant to thermally induced degradation, but degradation was seen in solutions stored under basic conditions. At 48 hours, approximately 50% of 5F-PB-22 was degraded to a single compound with a retention time 0.9min earlier than 5F-PB-22. By 96 hours, less than 1% of the original 5F-PB-22 remained. MS data indicate that the primary degradation product retains the alkyl-indole structural motif; however, an exact structure for this product remains to be determined. Similar results were seen for PB-22, with the exception that at 96 hours, approximately 10% of the original compound remained.

This research provides forensic scientists with noteworthy data on the continued rapid changes occurring with regard to synthetic cannabinoids and how the changes to avoid detection could result in the ingestion of unintended compounds.

Degradation, Synthetic Cannabinoids, Designer Drugs

A12 Fast Analysis of Heroin Using UHPLC

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After attending this presentation, attendees will become familiar with another rapid quantitative method using Ultra High Performance Liquid Chromatography (UHPLC).

This presentation will impact the forensic science community by introducing a faster and more efficient method for quantifying heroin using fast Liquid Chromatography (LC).

Heroin is one of the most commonly encountered illicit drugs in Singapore, making up 28% of exhibits analyzed by the laboratory. There are at least three types of heroin available in Singapore — South East Asian (SEA) Heroin No. 3, SEA Heroin No. 4, and South West Asian Heroin — each with a different chemical composition. In Singapore, SEA Heroin No. 3 is the predominant type used by drug abusers. Besides the active component diamorphine, adulterants such as caffeine and chloroquine are also frequently detected during analysis. Acetylcodeine and monoacetylmorphine are commonly encountered as by-products of the acetylation and hydrolysis processes, especially in low purity heroin (<5%).

Over the past ten years, the number of heroin exhibits seized by law enforcement agencies in Singapore has risen by about four fold. Besides the sheer volume of analysis required, the composition of the heroin encountered also poses challenges due to the presence of multiple components and late-eluting compounds in the exhibits. To cope with the escalating number of seizures and the pressure of short turnaround-times, the

current analytical method for the quantitation of diamorphine was further optimized in order to achieve an even higher throughput. Currently, the laboratory conducts quantitative analyses using Hypersil-5-ODS columns (20cm x 4.6mm, 5µm) on conventional High Performance Liquid Chromatography (HPLC) coupled with photodiode array UV detectors. This presentation explores the use of 2.7µm superficially porous particle column in order to achieve faster run times for heroin analysis. Superficially porous particle columns differ from the conventional fully porous particle columns as column efficiency is enhanced with smaller diffusion distances without the drawback of high back pressures. These columns also remove the need for tedious sample preparation usually required of sub-2µm particle columns, resulting in less sample preparation and improved productivity.

The objective of this presentation is to present a rapid quantitative method on UHPLC for analysis of heroin using a superficially porous particle column (Ascentis Express C18, 10cm x 4.6mm, 2.7µm). This study shows reduction in analysis times to approximately six minutes, which is an improvement in the efficiency of at least five-fold. The peak shape and resolution of the analytes were also improved. In addition, the robustness was enhanced, thus further ascertaining the performance of the new method.

Diamorphine, Fast LC, Method Development

A13 Evaluation of Human Scent As Possible Classification Evidence

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After attending this presentation, attendees will have expanded their understanding on how differences in the scent profile of individuals from different groups may be used to both identify and categorize them as part of such groups.

This presentation will impact the forensic science community by presenting a new and alternative way in which human scent evidence may be used to aid in the resolution of forensic cases. In addition, this presentation will describe the potential impact that the application of this new perspective on human scent can have on the criminal justice system and its processes.

To date, research studies within the forensic phenotyping field have attempted to predict physical characteristics of individuals through the use of different types of DNA analysis, with age being one of the focal points of these studies. A study published by Bocklandt et. al. in 2011 evaluated biological samples from different individuals in order to try to estimate their age by taking into consideration the variation in DNA methylation patterns with increasing age.¹ In another study published by Hannum et. al., a quantitative model of aging was evaluated in different human tissues and employed to measure the rate at which an individual's methylome changes with age.² Both of these studies represent some of the recent approaches that have been pursued to use genetic information to predict a subject's age. However, there are still some limitations to these types of DNA analysis that

have not been surpassed and that limit the application of these new techniques in the forensic field. Therefore, employing human scent evidence as a source of information that could provide insight on specific traits, like age, in an individual would provide an alternative tool when investigating the particular characteristics of an individual when physical evidence (e.g., blood or tissues) is not necessarily available. This would not only provide an additional way to characterize an individual but would also assist with categorizing individuals based on their shared traits. Applying this new perspective will make human scent a valuable asset in a forensic investigation and in the criminal justice system.

Past work evaluating human scent has shown that it can be used to differentiate individuals based on their characteristic Volatile Organic Compounds (VOCs), making it a useful form of forensic evidence.³ This has aided in the resolution of court cases by providing investigators with the ability to associate victims and offenders based on human scent left at crime scenes. On the other hand, other studies have focused on comparing body odor generated in different regions of the body, mostly as a way to define its source and nature. This study represents the first attempt to evaluate body odor in a way that specifically targets forensic applications. The present research investigated whether human scent can also be employed to classify individuals that share traits which contribute to their scent profiles and cause them to display similarities. Such similarities can be used to categorize individuals and exclude or include them as part of specific groups. For example, the determination of differences in body odor among individuals of different age groups may result in a new tool that can assist in crime investigations where the subjects may be quickly excluded or included as a potential suspect.

In this study, individuals from different age groups were recruited, their body odor was sampled, and their characteristic VOC profiles were analyzed by Headspace Solid Phase Microextraction Gas Chromatography/Mass Spectrometry (HS-SPME-GC/MS). Once collected, the profiles obtained were evaluated to determine which components provided information on how age could be used as a criterion for the categorization of individuals based on their VOC profile. This presentation will emphasize the way in which information obtained from the combination of features in a VOC profile could be employed to highlight links between individuals that share a specific trait—age. These results will not only enhance the amount of information that can be obtained during the evaluation of human scent, but also provide a broader perspective on how human scent may be used as a tool for classifying individuals into groups. In addition, these findings will also serve to complement the information obtained through other types of tests that are currently available and could be used for subject classification as well, such as DNA profiling.

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Human Scent, Volatile Organic Compounds, Subject Classification

A14 Hyperspectral Imaging of Latent Fingerprints and/or Palm Prints

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After attending this presentation, attendees will gain a new understanding about the usefulness of a combination of hyperspectral imagers with visible Continuous Wave (CW) lasers for detection of latent fingerprints and/or palm prints without pretreatment.

This presentation will impact the forensic science community by proposing a new strategy in criminal investigation. The proposed equipment can noninvasively and untouchably search and detect latent fingerprints/palm prints on a wall panel that are left as-is in a typical criminal investigation.

DNA profiling is considered to be the most reliable tool in the identification of individuals while fingerprints and/or palm prints are still one of the most important pieces of evidence in scientific crime detection because they also remain unchanged for life. It is important for comprehensive forensic examination to preserve residual epidermal cells, which are believed to include degraded DNA, in detecting latent fingerprints/palm prints in crime scenes. Optical detection has promise in addressing such situations because it makes noninvasive and untouchable detection possible if a wavelength that does not harm degraded DNA is selected. So far, hyperspectral imaging, so-called chemical imaging in the forensic science field, has been mainly evaluated for its potential to increase the sensitivity of detecting latent fingerprints treated by conventional methods.¹⁻³ In this report, the potential of hyperspectral imaging to detect untreated latent fingerprints/palm prints was examined.

A portable hyperspectral imager was developed. It could accumulate a Hyperspectral Data (HSD) cube of the view field of 30mm x 30mm consisting of 658 (line pixels) x 638 (scan pixels) x 248 (spectral pixels) within about 13 to 42 seconds depending on the exposure time. The originally designed spectral range was from 440nm to 1,000nm. The spatial and spectral resolutions were 46µm and 2.0nm, respectively. The distance from an object was automatically kept constant using a laser diode with an oscillation wavelength of 850nm and a photodiode. Illumination was provided by a light source with a 150W halogen lamp for reflectance measurements and by two kinds of CW lasers for static fluorescence measurements. The oscillation wavelength of each laser was, respectively, 460nm and 532nm and their output power was set to be 1.98W. Neither wavelength will harm degraded DNA. Appropriate low pass filters were used in fluorescence measurements.

The strategy was as follows: latent fingerprints/palm prints were looked for using either CW lasers and if they existed, their HSD were measured under the three kinds of illumination. Proper spectral images were reconstructed from the HSD. Latent fingerprints on wall panels made of steel sheets to the surface of which a polyethylene resin was applied and baked at 190-200°C were detected.

Ridge lines of a fingerprint or a palm print were clearly described in single-wavelength band images centered at 620nm with band width of 10nm under the CW laser excitations but not

so satisfactorily in the corresponding reflectance images under the white light illumination because of less ridge contrast. When two kinds of excitation laser wavelengths were compared, the 532nm excitation provided images of better quality. This was attributed to the fact that the background luminescence/fluorescence intensity was stronger at the 460nm excitation than at the 532nm excitation.

In conclusion, the present results suggest that the proposed strategy has promise to detect latent fingerprints/palm prints and, simultaneously, to preserve degraded DNA.

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Hyperspectral Imaging, Visible CW Lasers, Untreated Finger/ Palm Prints

A15 Detection of URB Series Synthetic Cannabinoids

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After attending this presentation, attendees will understand some of the difficulties associated with URB synthetic cannabinoid detection as well as certain advantages of the ability to detect these drugs.

This presentation will impact the forensic science community by delivering a possible detection method that can be implemented alongside a laboratory's current drug detection methods, while also serving as a basis for further research into this emerging class of drugs to allow for the development of better detection methods.

Use of the URB series is growing to replace synthetic cannabinoids made illegal by various pieces of federal and state legislation. The structural and pharmacological properties of each of the URBs prevent them from being scheduled by current federal legislation as a THC analog, and only a handful of states have enacted bans that cover the URBs. Some states, such as Kentucky, have enacted blanket bans that include all of the URBs while others have only explicitly banned some of the URBs. Their inclusion in products such as "Spice" requires that they be reliably detectable and identifiable by forensic drug laboratories.

Validated methods at the Kentucky State Police Eastern Regional Laboratory were initially used to determine under what parameters detection of URB cannabinoids would be optimal. These methods showed significant elution problems. A new method was developed with higher temperatures to improve elution, but degradation at these temperatures became its own problem. While the peaks became sharper as elution improved, breakdown products became evident and would also increase with prolonged time at room temperatures as opposed to nearly constant storage in a freezer. The developed method is adequate but not ideal.

Mock evidence was created and analyzed to determine the viability of this method under conditions that a drug analyst would most likely encounter. Each standard as well as a mixed sample

were applied to whole leaves and crushed leaf material, extracted, and analyzed. The leaf material showed no co-elution with any of the analytes as determined from method development analysis; however, detection was highly inconsistent across samples. Of the 16 mock evidence samples, only URB-602 was detected in a total of four samples.

As more synthetic cannabinoids are made illegal by state and federal legislation, there may be a rise in the use of the URB series. Further study in solving the encountered degradation problems, possibly through derivatization, could provide a reliable detection method that would reduce any lag time between legislation against the URBs and a laboratory's ability to detect them in evidence.

Detection, URB Series, Synthetic Cannabinoid

A16 Fluorescent Random Amplified Microsatellites (F-RAMS) Analysis of Mushrooms as a Forensic Investigative Tool

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After attending this presentation, attendees will understand how Fluorescent Random Amplified Microsatellites (F-RAMS) can be used to identify hallucinogenic mushrooms to species level and to obtain intra-specific variation within mushrooms — an attribute that can provide investigative leads in tracking the evidence source or associating criminal cases.

This presentation will impact the forensic science community by introducing F-RAMS, a DNA-based approach that could be used to provide investigative leads in criminal cases involving hallucinogenic mushrooms. Of additional interest is the potential adaptation of this "universal" method to any eukaryotic organism, when genome sequence information is limited.

In the illegal market, hallucinogenic mushrooms are seldom confiscated fresh or intact and, therefore, unambiguous morphological identification to species level is often not possible. Under most circumstances, the standard method for proving illegal distribution is chemical analysis of the indole alkaloid contents. However, chemical analysis cannot provide any species- and sub-species-specific information about the evidence that could aid forensic investigations in tracking the source or distribution networks. In contrast, a DNA-based approach can provide such information. Currently in molecular systematics, Internal Transcribed Spacer ribosomal DNA (ITS rDNA) sequencing is the most reliable approach for the fungal identification to species level.¹ However, DNA sequencing is not commonly used in many forensic laboratories where the profiling of microsatellites is the preferred method of choice. Moreover, the confined knowledge about fungal genomes poses a limitation in conventional microsatellite analysis.

In this study, the capability of a different DNA-based approach was tested. F-RAMS were used to profile mushroom evidence to species and sub-species level. The use of RAMS to study genetic variation within fungi was first described by Hantula

*et al.*² In this study, Hantula's approach was modified by adding a fluorescent label to the RAMS primers for profiling of mushroom species, thus allowing for a more sensitive amplicon separation through capillary electrophoresis. Thirty-seven hallucinogenic and non-hallucinogenic mushrooms of the genera *Amanita* and *Psilocybe*, including 15 samples of the species *Amanita rubescens* were profiled using two fluorescently labeled degenerate primers, 5' DD (CCA)₅ and 5' DHB (CGA)₅, which target different microsatellite repeat regions. The amplicons were separated using an ABI 310 Genetic Analyzer and analyzed using the Genemapper® software version 4.0.

Among the two primers, 5' DHB (CGA)₅ provided more reliable data for identification purposes by grouping samples of the same species and clustering closely related species together in a dendrogram based on amplicon similarities, computed using the Jaccard similarity coefficient. A high degree of intra-specific variation between the 15 *A. rubescens* samples was shown with both primers and the amplicons generated for all *A. rubescens* samples were organized into three classes of amplicons (discriminant, private, and marker) based on their individualizing potential.

The data demonstrated that F-RAMS analysis of mushrooms is a fast and reproducible method and that F-RAMS has the potential to become a powerful molecular forensic tool to complement chemical analysis. RAMS is a "universal" method in that it does not require prior knowledge of the genome being profiled and could be potentially adapted to any eukaryotic organism when genome sequence information is limited.

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Hallucinogenic Mushrooms, Profiling, RAMS

A17 Analysis of Trace Organics at Parts-per-Trillion Level Concentrations Using a Novel Sample Preparation Technique: Fabric Phase Sorptive Extraction

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After attending this presentation, attendees will understand the working principle, advantages, and potential applications of the recently developed sample preparation technique known as Fabric Phase Sorptive Extraction (FPSE) in preparing forensic samples for instrumental analysis.

This presentation will impact the forensic science community as well as scientists interested in analyzing trace organic analytes in food, pharmaceutical, biological, environmental, and toxicological samples of forensic significance by providing a simpler, inexpensive sample preparation technique capable of handling original samples without any matrix clean-up which is equally effective both in the laboratory and in the field.

FPSE has shown to be highly effective in extracting illicit drugs, nitroaromatic explosives, and other trace organic compounds of forensic significance directly from biological and environmental

samples. The current study illustrates the application of FPSE in extracting Polycyclic Aromatic Hydrocarbons (PAHs) from environmental water. The United States Environmental Protection Agency (USEPA) has included 16 PAHs in the EPA Priority Pollutants List. Many of these PAHs are known or suspected carcinogens. PAHs enter the environment via anthropogenic and natural processes. Due to their detrimental effect on human health and public safety, large numbers of analytical methodologies have been developed over the years including Solid Phase Extraction (SPE), Liquid-Liquid Extraction (LLE), solid-phase microextraction (SPME), Stir Bar Sorptive Extraction (SBSE), purge-and-trap, and others. Due to the complexity of environmental water as a sample matrix (high content of particles, plant debris, microbes, dissolved organic materials), most of the samples require sample clean-up prior to the extraction of target analyte(s) (e.g., centrifugation and filtration). Due to the high lipophilicity of PAHs, they tend to be absorbed on surfaces. As such, conventional multi-step sample preparation approaches may potentially cause a high degree of PAH loss during different sample preparation steps. Consequently, there is a high demand of a sample preparation approach that is capable of pre-concentrating the target analyte(s) directly from the original sample without any matrix clean-up and does not require solvent evaporation and sample reconstitution. FPSE, recently developed at the International Forensic Research Institute, has addressed most of the inherent shortcomings of conventional extraction and microextraction techniques. FPSE utilizes a small flexible media coated with sol-gel hybrid organic-inorganic polymeric material as the extraction sorbent. The sol-gel coating process results in a highly porous polymeric network chemically bonded to the substrate surface. The hybrid material inherently possesses high thermal, chemical, and mechanical resistance as well as high specific surface area. Compared to the Primary Contact Surface Area (PCSA) of SPME and SBSE, FPSE has a large advantage (1000mm² for FPSE vs. 100mm² for SBSE vs. 20mm² for SPME). The high PCSA of FPSE media increases the probability of the successful sorbent-analyte interaction for an effective analyte extraction, resulting in fast extraction equilibria along with a high preconcentration factor. Unlike SPE, SPME, and SBSE, there is no limitation in solvent selection for the back-extraction (for solvent mediated desorption) and a judicious solvent selection allows injecting the same sample in both GC and HPLC systems.

Another unique feature of FPSE is its ability to perform the whole sample preparation exercise *in situ* in the close proximity of the sample source which significantly reduces the laborious and expensive processes of collecting the sample from the field, transporting it to the analytical laboratory, and storing it under controlled temperature before the sample preparation for instrumental analysis begins.

The current study consisted of FPSE on water samples containing four of the 16 EPA PAH pollutants: anthracene, fluoranthene, fluorene, and phenanthrene. The effects of extraction and desorption parameters such as extraction time, ionic strength, stirring rate, desorption time, and desorption solvent system on the extraction/desorption efficiency were investigated and optimized. Coupled to high performance liquid chromatography with a UV detector, the optimized FPSE method was applied to the analysis of four PAHs in aqueous samples at 200 parts-per-trillion level concentrations. The analytical figures of merit including linearity, Limit Of Detection (LOD), Limit Of Quantitation (LOQ), and reproducibility surpass the respective values reported in the literature for other techniques such as SBSE and SPME.

This is purportedly the first report of the direct extraction of PAHs at 200 ppt level concentration in solventless microextraction and analysis using HPLC-UV.

PAHs, FPSE, Environmental Forensics

A18 Latent Print Development on Adhesive Side of Dark Colored Tape Using Ash Gray Powder with Liqui-Nox® Detergent

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After attending this presentation, attendees will learn why Liqui-Nox® detergent was determined to be a viable replacement for Photo-Flo® 600 Solution in the Ash Gray Powder solution used to develop latent prints on the adhesive side of dark colored tape at the FBI Laboratory. Ash Gray Powder mixed with 50:50 Liqui-Nox®:water solution (experimental) was expected to work in the same manner as Ash Gray Powder mixed with Photo-Flo® 600 Solution (control) because both chemicals are detergents. Therefore, it was hypothesized that Liqui-Nox® would be an adequate replacement for Photo-Flo® 600 Solution.

This presentation will impact the forensic science community by dealing with latent print development, particularly of interest for the users of the Ash Gray powder process. Photo-Flo® 600 Solution is used for several different processing techniques, including Sticky-side powder, Amido Black (Fischer 98), and Amido Black (water base).¹ Photo-Flo® 600 Solution is currently used as the detergent in the Ash Gray powder solution to develop latent prints on adhesive side of dark colored tape; however, Kodak®, the company that manufactured Photo-Flo® 600 Solution, has sold its document imaging and personal imaging businesses, ceasing production of many of its chemicals.² A comparable replacement detergent is needed for the Ash Gray processing technique.

Laboratory-controlled latent prints were deposited on the adhesive side of 100 pieces of black electrical tape (measuring approximately three inches each). Participants groomed fingers by rubbing across nose and forehead to include sebaceous and eccrine secretions. Prints were deposited and the process was repeated. Tape pieces with prints were cut down the middle vertically. Each half of the same print was processed using Ash Gray Powder mixed with either Photo Flo® 600 Solution or 50:50 Liqui-Nox®:water solution. Randomization was employed to select which half would be processed with the control and which half with the experimental Ash Gray Powder solution. Both solutions were applied with a camel hair brush and were left on the adhesive side of tape for 60 seconds before rinsing with tap water. Scanned images of the tape halves were analyzed for value and quality. A qualified latent print examiner evaluated the development of each latent print half and the quality of the resultant prints. A separate qualified latent print examiner analyzed the two processes by comparing and assessing the value for identification of the individual print halves. Another set of images (non blind) in which all experimental documentation was provided was used for statistical analysis.

The results of this study validated using Liqui-Nox® detergent, instead of Photo-Flo® 600 Solution as a replacement for the Ash Gray Powder solution to develop latent prints on the adhesive side of dark colored tape. For this specific purpose, Liqui-Nox® worked reliably to develop latent prints. This study would not necessarily deem Liqui-Nox® as a viable replacement in all processing methods that involve the chemical Photo-Flo® 600 Solution, rather only as a viable replacement in the Ash Gray adhesive processing method.

References:

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2. Daneman, Matthew, (2013), "Kodak gets court OK to sell film business," *USA Today*, 6/20/13

Latent Prints, Tape Processing, Ash Gray Powder

A19 GC/MS and GC/IR Analysis of Methcathinone Analogs

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After attending this presentation, attendees will understand the difficulty of analyzing methcathinones via Gas Chromatography/Mass Spectrometry (GC/MS) and how Gas Chromatography-Infrared Spectroscopy (GC-IR) can be used to overcome this problem. Attendees will also learn the settings used to obtain the spectra for the drugs as well as the characteristics of each spectrum.

This presentation will impact the forensic science community by providing a method that can be used to analyze methcathinones as well as reference spectra to aid in identification of these drugs.

Methcathinone is a psychoactive stimulant, structurally similar to methamphetamine. Methcathinone itself is a Schedule 1 drug, which has led to the production of many methcathinone analogs that fall outside of the criteria described by the legal system. The majority of the analogs manufactured are altered in such a way that the compound largely remains the same, but with a minor difference that does not significantly change the effects. The methcathinone structure is modified at three positions to produce these analogs including: the alkyl side chain, the amino group, and the aromatic ring. Substitutions on the aromatic ring are the most diverse and have included methyl, fluoro, methoxy, and methylenedioxy groups. The most common substitution on the amino group is a pyrrolidine ring, while the aliphatic side chain is commonly modified by increasing the length of the carbon chain.

The legal status of these analogs is problematic for drug analysts. Only three analogs have been placed into Schedule 1 of the Controlled Substances by the DEA: methyldone, mephedrone (4-methylmethcathinone), and methylenedioxypropylvalerone (MDPV). The control of these analogs varies by state. With their use on the rise and the unclear scheduling, laboratories are tasked with correctly identifying these analogs.

GC/MS is the gold standard in drug identification; however, its use is limited in the analysis of methcathinone analogs. GC/MS can help the analyst by providing information about where a modification has been made. A compound with a pyrrolidine ring on the amino group and no modification to the aliphatic side chain are indicated by a base peak at 98m/z. A compound where the only modification is on the aromatic ring shows a base peak at 58m/z. However, GC-MS analysis cannot differentiate between positional isomers due to their nearly identical mass spectra. The use of GC-IR can reliably distinguish between these isomers.

Both GC/MS and GC-IR data will be presented to demonstrate the usefulness of GC-IR in methcathinone identification compared to GC/MS. The following drugs will be presented: 3 and 4-fluoromethcathinone; 2, 3, and 4-methylmethcathinone; 2, 3, and 4-methoxymethcathinone; α -pyrrolidinopropiophenone (PPP); 2, 3, and 4-methyl- α -PPP; 4-methoxy- α -PPP; 3,4-methylenedioxy- α -PPP; 3,4-dimethylmethcathinone; 2,3-methylenedioxy-methcathinone; butylone; pentadone; methyldone; and MDPV.

Methcathinones, GC-IR, GC/MS

A20 Development of an Identification and Derivatization Method for Synthetic Cathinones by GC/MS Using Perfluoroacyl Anhydrides

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After attending this presentation, attendees will have a better understanding of the use of perfluoroacyl anhydrides as derivatizing agents for compounds from the synthetic cathinone family.

This presentation will impact the forensic science community by providing a simple method for the identification of synthetic cathinones using perfluoroacyl anhydrides as a derivatizing agent and Gas Chromatography-Mass Spectrometry (GC/MS) for analysis.

Synthetic cathinones, more commonly referred to as "bath salts," are becoming increasingly abused in the United States. As a result, forensic drug laboratories are receiving an increasing number of samples believed to contain these compounds. Synthetic cathinones are gaining popularity due to their psychoactive and stimulant properties, which are similar to those of amphetamine and cocaine. They give a pleasurable rush and increase sociability; however, they can have severe negative side effects that put the health of the public at risk. Therefore, the DEA has placed three synthetic cathinones in Schedule I of the Controlled Substances Act: mephedrone, methylone, and MDPV. Several other cathinones have been recently controlled under the Control Substances Analogue Act.

A drug is scheduled based on its exact structure; therefore, the alteration of a single position in the molecule could yield a new, legal drug that would theoretically have the same effects as the original scheduled drug. However, the Federal Analog Act of 1986 states that any compound that is "substantially similar," both in structure and pharmacological effect, to a Schedule I or II controlled substance and intended for human consumption is to be treated as if it were also scheduled. Because these are synthetic compounds, their molecular structure can be slightly altered to circumvent scheduling under the DEA's Controlled Substances Act.

Due to the minor structural variations and legal ramifications associated with synthetic cathinones, identification of the exact structure is imperative. The three scheduled compounds each have the potential for positional isomers, which result in ambiguous mass spectra when analyzed by GC/MS. Because GC/MS is the primary method of drug identification used in a forensic laboratory, it is essential to develop a simple method that makes differentiation as straightforward a process as possible. Derivatization has proven useful in the past for determining differences in mass spectral results when underivatized samples resulted in analogous mass spectra.

Three perfluoroacyl anhydrides were tested as derivatization agents for the synthetic cathinones: Trifluoroacetic Anhydride (TFAA), Heptafluorobutyric Anhydride (HFBA), and Perfluoropropionic Anhydride (PFPA). The ability of these anhydrides to derivatize compounds for successful differentiation by GC-MS was determined using 11 synthetic cathinones: 2-fluoromethcathinone, 3-fluoromethcathinone, 4-fluoromethcathinone, 2-methoxymethcathinone, 3-methoxymethcathinone,

4-methoxymethcathinone (methedrone), 2-methylmethcathinone, 3-methylmethcathinone, 4-methylmethcathinone (mephedrone), 2,3-methylenedioxy-methcathinone, and 3,4-methylenedioxy-methcathinone (methylone).

All three anhydrides were successful in giving differentiation among the mass spectra of the positional isomers. The addition of the anhydride to the amine functional group generated a difference in the relative abundance of the major ions between the positional isomers and occasionally gave one or two different major ions between the isomers. In the majority of the cases, when the derivatized isomers were placed in a mixture, they were easily distinguished both by retention time and the mass spectral results, both of which matched the results from running the compounds on their own.

When TFAA, HFBA, or PFPA are used as derivatizing agents for the synthetic cathinones, an increase in complexity is found in the mass spectra. This complexity creates individual mass spectra that are sufficient for both identification and differentiation.

Synthetic Cathinones, Derivatization, GC/MS

A21 Enhancing the Value of Information Derived From the Measured Diameters of Bullet Holes

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After attending this presentation, attendees will understand the relationship between bullet diameter and the size of the bullet hole in common materials found at shooting incidents.

This presentation will impact the forensic science community by providing information about the relationship between the diameter of a bullet and the factors affecting the diameter of the hole it produces in various materials, which can then be used to aid in shooting reconstructions.

In the last century, forensic firearms analysis has become increasingly scientific with technological developments such as the comparison microscope for the comparison of fired bullets and/or expended cartridge cases and computer-aided assessment of markings on such bullets and casings. These methods are useful as long as one has a suspect, gun, and/or spent round and casing to make comparisons; however, there are criminal incidents where neither the bullet nor the gun is recovered. It is also possible to have a scene with many guns involved with many bullets present and the question arises as to which firearm was used to produce which hole. In these cases, a comparison microscope or computer search would not be able to identify a linkage between the hole and the firearm. If the primary evidence in the crime scene is the bullet hole, can a suspect firearm be eliminated? What can be learned from the hole at the impact site about the projectile that produced it? This research sought to provide answers to these questions by determining the relationship between the diameter of the fired bullet and the size of the impact hole in common materials and also statistically assessing whether the size of the bullet hole could be used to identify bullet diameter.

This experiment involved shooting at a variety of common materials including gypsum drywall, wood, metal, plastic, and vinyl. A range of bullet diameters, from .22 to .45 caliber, were tested using a diverse selection of bullet sizes and designs including full

metal jackets, lead round nose, jacketed hollow points, and semi-jacketed hollow points. Multiple firearms and cartridges were used to ensure that holes were made with a representative range of projectile masses and velocities. The data were then statistically analyzed to evaluate the validity and limitations of estimating the bullet diameter from the bullet hole in various materials. The results of this research showed that although the size of the bullet hole varied based on the impact material, more information about the bullet diameter can be ascertained from the bullet hole than is currently being used in crime scene reconstructions. The conclusions obtained from this research will serve to improve the amount of information that can be derived from shooting events and thus provide more accurate information for reconstruction.

Reconstruction, Bullet Hole, Bullet Diameter

A22 Mobile Crime Scene Applications: An Evaluation of Their Use and Future Direction

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The goals of this presentation are to introduce attendees to a mobile crime scene application (app) by detailing some of an app's features, offer an evaluation of the app when used in a real-world setting, and propose modifications and additions that could be incorporated into future updates or a new application altogether.

This presentation will impact the forensic science community by introducing the emerging concept of using electronic tablets in the field to record information.

With many forensic science disciplines coming under scrutiny in recent years, it has become the objective of the forensic community to improve and standardize as many of the current practices as possible. Since crime scene investigation is a subjective field, it stands to face strong criticism in the courtroom. It is also becoming one of the slower steps in the analysis process; while DNA and chemistry technologies are getting faster and faster, the human role in analysis is more difficult to expedite. There is also disparity in evidence collection and reporting methods among agencies. Emerging technologies should therefore focus on reducing the time spent documenting a scene and streamlining and standardizing the documentation and collection process. To be effective, a technology should also be straightforward to learn and implement as well as cost effective.

Data was collected from a survey given to professionals in the field of crime scene investigation as well as an evaluation of a mobile crime scene app's performance, considering the aforementioned criteria, while documenting mock crime scenes. Experts were questioned regarding their current approaches to scene documentation and reporting as well as their preferences and technological proficiencies. The goal of the survey was to establish a discrepancy between digital natives and digital immigrants and how this factors into how the ease of learning of the software is evaluated. After recording mock crime scenes, the app was qualitatively assessed with respect to each established criteria. In its current state, the app will not supplant any existing field instruments, with the exception of paper and stencils. While incorporating photos in one's field notes is convenient, the iPad's® camera cannot be substituted for the traditional digital single lens reflex cameras commonly used in crime scene investigation. If the

department has to purchase the equipment and the app, then it may be cost prohibitive. However, if the app reduces the amount of time the crime scene investigator spends documenting each scene, then reduced labor costs may in time offset equipment costs. It was recognized that while the app provided streamlining of case notes, many alterations and/or additions could be made to create a better crime scene documentation tool. Recommendations for these modifications will be presented. It is hoped that future studies with this application will analyze the use of this app in more real-world settings and in the hands of working crime scene investigators. There are other applications on the market that also claim to improve note-taking and documentation; future studies will compare and contrast these apps with the one evaluated here.

Investigation, Standardization, Tablet

A23 Preparative Approaches to the Isolation and Purification of Kanna Alkaloids: Mesembrine, Mesembranol, Mesembrenone, Hordenine, Sceletenone, and Sceletium A4

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After attending this presentation, attendees will know the alkaloids present in the plant kanna (*Sceletium tortuosum*) and will be able to describe a method to separate and purify mesembrine, mesembranol, mesembrenone, hordenine, sceletenone, and sceletium A4 from plant products.

This presentation will impact the forensic community by raising awareness of this plant and its associated alkaloids, which may have abuse potential, by providing a method for its analysis, and by developing a method for preparing reference standards, which may be used in a laboratory to confirm the identity of kanna-derived products.

Kanna, a flowering cactus from southern Africa, is known to contain psychoactive alkaloids, including mesembrine, mesembranol, mesembrenone, sceletenone, sceletium A4, and hordenine. It is currently legally sold online and in some health shops in the United States as a homeopathic remedy or a natural high. With increasing popularity in the U.S., crime laboratories are more likely to encounter kanna in seized material, making it essential to have standard reference materials which meet the Scientific Working Group for the Analysis of Seized Drugs (SWGDRUG) criteria, either by synthesis or isolation.

This project's objective was to develop usable standards of the six main alkaloids in kanna. The goal of this project was to isolate and purify the alkaloids from the kanna plant powder through chromatographic means that would be readily available to most crime laboratories.

An initial crude separation of the alkaloids was done by extracting the kanna powder in acetonitrile and then spotting that extract onto a Thin Layer Chromatography (TLC) plate. After scraping the bands and reconstituting them in acetonitrile, the Gas Chromatography/Mass Spectroscopy (GC/MS) analysis of the band sections proved that the alkaloids have the potential to be separated and purified. A Selective Ion Monitoring (SIM) GC/MS method was developed to allow for easier and faster separation and identification of the alkaloids. The oven temperature increased

from 70°C to 325°C during the 18.25-minute run. The full scan method was used to look for all ions between 44 and 500m/z. The SIM method isolates 58, 70, 77, 96, 121, 175, 218, 219, 243, 248, 266, 274, 287, 289, 290, 309, 324m/z because this list contains the three most distinguishing mass fragments for each alkaloid.

An acid/base extraction with ethyl acetate as the organic solvent was developed to extract all of the alkaloids with the greatest efficiency. The SIM Liquid Chromatograph/Mass Spectrometer (LC/MS) method detected the M+1 ions for each of the alkaloids. An isocratic eight-minute run with the solvent 0.1% TFA in water, acetonitrile, and isopropanol (80:10:10) flowed through the Agilent® Eclipse Plus C18 column at 0.6 milliliters per minute. This method was developed to for two reasons. One was to determine if the alkaloids could be separated and collected from an analytical scale chromatographic method. The second reason was to monitor the contents of the various fractions produced during the subsequent preparative steps. Sceletenone, sceletium A4, and hordenine were substantially resolved to allow for fraction collection from an analytical LC/MS method.

Preparative separation of the compounds was attempted using a two-dimensional separation scheme starting with normal phase preparative column chromatography via a Teledyne ISCO CombiFlash® Companion® with a dichloromethane and 10% triethylamine in methanol gradient. This was followed by reverse phase preparative chromatography of the step one isolates via the same CombiFlash® Companion® with a gradient between 0.1% trifluoroacetic acid (TFA) in acetonitrile and 0.1% TFA in HPLC grade water. This two-dimensional chromatographic separation system failed to achieve the necessary resolution of the components of the extract needed for standard reference materials. The similarity in structure of mesembrine, mesembranol, and mesembrenone, the small concentrations of hordenine, sceletenone, and sceletium A4, and the fact that hordenine is a fairly small compound all contributed to the challenges experienced while trying to isolate these six alkaloids. Further work to optimize the separation and scale it up to a preparative procedure will be the subject of future research.

Sceletium Tortuosum, Kanna, Alkaloid

A24 Improving IR Searches in the PDQ Database to Enhance Investigative Lead Information From Automotive Paints

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After attending this presentation, attendees will understand the principles of pattern recognition and data fusion techniques, advantages of preprocessing infrared spectra using wavelets, and the importance of using search prefilters in library searches to improve the accuracy of infrared spectral searches in the Paint Data Query (PDQ) database.

This presentation will impact the forensic scientific community by demonstrating the advantages of using multivariate statistical and signal processing techniques to enhance current approaches to data interpretation of forensic paint examinations and to aid in evidential significance assessment, both at the investigative lead stage and at the courtroom testimony stage. There is also potential for direct impact with the 75 local, state, and federal forensic laboratories currently using the PDQ database in

the United States as well as international forensic laboratories that utilize PDQ including: the Forensic Laboratory Services Division of the RCMP, the Centre of Forensic Sciences in Toronto, Canada, the ENFSI network of European forensic science institutes, the Australian Police Services, and the New Zealand Police Services.

Modern automotive paints have a thin color coat which, on a microscopic fragment, is often too thin to obtain reliable chemical information. The small size of the fragment also makes it difficult to compare it with the manufacturer's paint color standards. Fortunately, adhesion between paint layers is usually very strong and so both primer layers are often transferred during a collision if the clear coat and color coat layers are also transferred. As the primer layers and clear coat layer are usually unique to the automotive assembly plant where these layers were applied, combining chemical information obtained from the Fourier Transform Infrared (FTIR) spectra of the two primer layers and from the clear coat layer has made it possible to rapidly and accurately identify the assembly plant of an automobile from its paint system.

Applying data fusion techniques where spectra from multiple sources (e.g., IR spectra of the clear coat and primer paint layers) are combined and class membership information is extracted, search prefilters for the PDQ library have been developed to determine the model and line of an automotive vehicle from which an unknown paint sample originated. Even in challenging trials using a database of 380 paint samples where the clear coat and undercoat layers evaluated were all the same make with a limited production year range (2000-2006), the respective assembly plants were correctly identified using only chemical information extracted from FTIR spectra of the clear coat and primer paint layers due to a synergistic effect that occurred when spectra of the different layers of paint were fused. Applying wavelets to preprocess the IR spectra of the clear coats and primer layers, advantages associated with higher order data fusion techniques were achieved when a genetic algorithm for pattern recognition and feature selection was used to identify the informative wavelet coefficients in the fused data.

The development of search prefilters for the PDQ database that utilize multiple spectra is needed to extract investigative lead information from clear coat and primer layer paint smears. Searches performed using the current PDQ database tend to generate a large number of hits because the chemical information in the current database is described only in terms of generic chemical formulations (e.g., acrylic melamine styrene or acrylic melamine styrene polyurethane) to delineate the chemical composition of clear coats. Due to the limited number of formulations used to prepare clear coats, the use of additional information in the search improves both the selectivity and accuracy of the searches that involve clear coats. An added advantage of a pattern recognition-assisted approach to identify paint samples is an increase in the accuracy of the search because all FTIR spectra in the database are searched. The use of the search prefilters gives fewer hits, which translates into a significant time savings for the forensic scientist. Information derived from these pattern recognition searches can also serve to quantify the general discrimination power of original automotive paint comparisons encountered in casework and will further efforts to succinctly communicate the significance of the evidence to the courts.

Pattern Recognition, Data Fusion, Library Searching

A25 Forensic Analysis of Dyed Fibers Via Time-of-Flight Secondary Ion Mass Spectrometry

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After attending this presentation, attendees will discover how Time-of-Flight Secondary Ion Mass Spectrometry (TOF/SIMS) can be used to obtain accurate mass spectra and mass spectral imaging (i.e., chemical imaging of the longitudinal surface and/or cross-sectional area of dyed fibers) and how these data, when coupled with an analytical dye database, can be used to unambiguously identify commercial textile dyes.

This presentation will impact the forensic science community by providing an improved high-precision method of dyed fiber analysis compared to current standard fiber analysis methods.

This research was based on the hypothesis that method development using TOF/SIMS would provide an advanced dyed fiber analysis protocol that: (1) is a relatively rapid, direct, and unambiguous tool for the identification of commercial disperse and acid dyes on or in synthetic fibers; (2) is effective at relatively low concentrations of dye in the fiber and can be applied to the cross section of a single fiber; (3) enables location-dependent, two-dimensional, mass spectral-based chemical imaging of dyed fibers on the fiber surface or in the fiber cross section; (4) enables dye analysis with minimal destruction of fiber; (5) enables surface "cleaning" in the event of surface contamination or of dye degradation or loss to enhance spectral resolution, and, (6) can be combined with a comprehensive database to provide immediate unambiguous dye identification.

The results presented support the hypothesis and have potential to positively impact the forensic trace evidence community by providing an improved high-precision method of dyed fiber analysis compared to current standard fiber analysis methods. Specifically, TOF/SIMS was employed to identify various dyes in finished textile fibers from both the fiber surface and, with the aid of cryomicrotomy, fiber cross section. Dyed fibers analyzed include disperse dyes in polyester and acid dyes in nylon 6.6. Results obtained by TOF/SIMS were consistent with analyses using Liquid Chromatography TOF Mass Spectrometry (LC/TOF/MS). Using C.I. Acid Blue 25 as an example, a method has been developed to detect acid dyes with only 0.1% dye by weight of fiber on nylon 6.6 fiber surfaces and cross sections. In this method, a C_{60}^+ ion beam was used as a "soft cleaning" method to remove surface contamination and any Bi ion beam-induced damaged materials. This method enabled increased molecular ion signal integrated intensity, which results in improvement in signal-to-noise and, thus, mass spectral resolution of the dye. An example showing the identification of dyes from one single fiber cross section will be presented to demonstrate that positive identification is feasible with minimal destruction of fiber trace evidence.

Currently, no widely available comprehensive analytical database of dye structures exists to support the accurate analysis of dyes within textile fibers. In addition, most dye identification

methods require extraction of dye from fiber in order to obtain mass spectral and other key analytical data. Research underway to provide a comprehensive mass spectral dye database using LC/TOF will be briefly reviewed. This database, when combined with the TOF/SIMS method, provides a powerful combination of mass spectral dye analysis. In addition, the wide scope of TOF/SIMS provides both elemental and mass spectral analyses for identification of a wide variety of non-colored chemical compounds on or in fibers, as well as expansion of the method to forensic analysis other materials, such as surface coatings (paints, lacquers), plastics, composites, inorganics, and metals.

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TOF/SIMS, Dyed Fibers, Trace Evidence

A26 Fingerprint Submission Decision Making Within a United Kingdom Fingerprint Laboratory: Do Experts Get What They Need?

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After attending this presentation, attendees will gain insight into the decision-making processes involved in assessing which pieces of developed friction ridge detail should be submitted to fingerprint experts to be considered as part of the Analysis, Comparison, Evaluation-Verification (ACE-V) process through the presentation of a United Kingdom-based study.

This presentation will impact the forensic science community by increasing the transparency of the fingerprint submission decision-making processes by providing a case example which demonstrates the importance of carrying out quality assurance in this area and highlights the need to understand the differing ridge detail sufficiency thresholds of fingerprint laboratory staff and fingerprint experts so as to maximize the resource efficiency of fingerprint evidence.

In the United Kingdom, items of forensic evidence are routinely submitted to a police force in-house fingerprint recovery laboratory where physical and chemical techniques are utilized to visualize latent fingerprints. It is then the responsibility of fingerprint laboratory staff to examine the areas of developed friction ridge detail on the item and to determine which are of sufficient quality to be submitted to fingerprint experts for comparison with suspect fingerprints. Fingerprint laboratory practitioners are either trained in-house by police forces or externally by the National Policing Improvement Agency, with the focus of this training on selecting and carrying out chemical fingerprint development techniques, not on the recognition selection and analysis of friction ridge detail. In contrast, fingerprint experts carry out a five-year training program that focuses on the ACE-V process. In the domain of psychology, the differences in ability in pattern recognition between "experts" and "novices" has been well discussed and experts have been found to excel at the detection and recognition of patterns.

In recent years, forensic science practice has been criticized for not upholding sufficiently scientific standards. In the United Kingdom, the appointment of the Forensic Science Regulator, tasked with ensuring that the provision of forensic science services is subject to appropriate scientific quality standards, has arguably paved the way for change. Fingerprint recovery laboratories must now achieve accreditation to the international standard ISO17025 which has led to an increased reliance on the use of Home Office-recommended fingerprint development techniques that have been United Kingdom Accreditation Service accredited, and the adoption of quality assurance procedures in relation to these techniques. However, the decision to submit an area of friction ridge detail to a fingerprint expert or to discard the detail does not fall directly under these quality assurance procedures or Home Office-based accreditation.

This present study looked to further the understanding of the effectiveness of the fingerprint laboratory decision-making processes within the United Kingdom Metropolitan Police Evidence Recovery Fingerprint Laboratory. Metropolitan police fingerprint experts were asked to identify a series of partial fingerprints to ten print cards provided. A combination of the resultant identifiable and insufficient fingerprints was selected to form the experimental set. These fingerprints were given to fingerprint laboratory practitioners who stated whether or not they would (mark up) and submit each mark to fingerprint experts. The assessments of the expert and laboratory staff were compared to identify incidences in which experts were given marks of too poor a quality or identifiable marks were discarded by the laboratory.

This presentation demonstrates the differences in the submission thresholds between the fingerprint laboratory and the fingerprint experts and the effect of context in the form of the crime category on this decision. It highlights the importance of continued quality assurance work in this arguably subjective area and the need to focus training and policy on improving the efficiency of the submission of ridge detail to fingerprint experts by the fingerprint laboratory to ensure that experts "get what they need."

Fingerprint, Decision Making, Interpretation Thresholds

A27 Quantifying Degradation of Explosives in a Saline Environment

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After attending this presentation, attendees will be aware that explosives, namely TNT and RDX, are affected by a saline environment. Details on the quantified decomposition of the compounds and degradation products formed will be discussed. Furthermore, the effects of other environmental conditions (such as UV exposure and temperature) will also be discussed.

This presentation will impact the forensic science community by providing a better understanding as to what effects a saltwater environment may have on the detection of explosives. Further, it will provide a method for the analysis of explosives in aqueous solutions using a number of different analytical techniques, as well as a way to quantify the concentration of explosives in solution. The forensic science community will also be provided with identified degradation products which could aid in the detection of a decomposed sample.

The degradation of explosives in the environment is important for a variety of reasons including remediation, post-blast detection, and evidence storage. While a number of studies have been completed that have determined how explosives degrade in soil and in wastewater, a comprehensive study of the

effects of a saline environment in explosive degradation has not been completed. Studying explosives degradation in a saline environment is critical for forensic cases involving a marine setting, such as the USS Cole bombing.

In this study, explosives (TNT and RDX) were dissolved into saline solutions ranging from no salinity to twice the salinity of ocean water and quantified based on exposure time. In addition, other environmental factors such as a combination of salinity and UV exposure as well as salinity and heat were studied. Analysis of the samples involved a variety of analytical techniques to allow for the development of the most practical and information-rich methods. Analytical techniques used include: Gas Chromatography Negative Chemical Ionization Mass Spectrometry (GC/NCI/MS), Direct Analysis in Real Time Mass Spectrometry (DART®/MS), Liquid Chromatography Mass Spectrometry (LC/MS-MS), UV-Vis spectroscopy, and Ion Mobility Spectrometry (IMS).

Results detail the developed optimized sample preparation and analyte identification and quantification methodologies. Extraction efficiencies using a number of different organic solvents were determined to find the optimal solvents to use for TNT and RDX. Sample quantification was performed using a 1200 Varian GC/MS by co-injecting the sample (1 µL) with an isotopically labeled standard (1 µL). The instrument was operated in negative chemical ionization mode for improved detection sensitivity of the analytes. Each explosive required the development of optimized experimental conditions such as an injection temperature, column oven ramp, and flow rate. Using these developed methods, it was found that the degradation of TNT in a saline environment begins within the first month of storage. However, if salinity exposure is coupled with UV light exposure, this degradation is significantly sped up, with nearly the entire sample being degraded in a matter of days. RDX has been shown to be more stable, with little degradation occurring in the first three months except when exposed to UV light. Additionally, studies have been completed using DART®/MS and LC/MS/MS to aid in the identification of the degradation products. Current studies are looking into the identification of the degradation products and potential mechanisms of degradation as well as the effects of the combination of heat and salinity on degradation.

Explosives, GC/MS, DART®

A28 Increasing the Sensitivity of Mass Spectrometric Analysis of Nuclear Materials

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After attending this presentation, attendees will understand how mass spectrometry can provide the nuclear forensics community with high accuracy and precision measurements, specifically showing how laser postionization can be used to increase the overall sensitivity of mass spectrometric analysis of nuclear materials.

This presentation will impact the forensic science community by advancing the state-of-the-art mass spectrometric analysis of nuclear materials to directly support nuclear nonproliferation and other national security missions.

This presentation will focus on current state-of-the-art mass spectrometry methods used for non-proliferation and nuclear forensics analyses. The efficacy of these techniques is reliant on the ability to detect trace elemental signatures from complex sample matrices. The use of laser-induced postionization to enhance the

sensitivity of mass spectrometric analyses will be demonstrated.

Mass spectrometric methods (e.g., Thermal Ionization Mass Spectrometry (TIMS), Inductively Coupled Plasma Mass Spectrometry (ICP-MS), and Secondary Ion Mass Spectrometry (SIMS)) are limited by their ionization efficiency which is simply the number of ions detected divided by the amount of material analyzed. Typically, ionization efficiencies for these methods fall in the 1-2% range. This means that 99-98% of the material is neutral (in ground and excited states) and thus can be accessed by photoionization schemes using high-energy, focused-laser beams. Postionization (laser-induced ionization of desorbed neutrals) is applicable to most, if not all, mass spectrometry methods currently employed for nuclear forensic analyses. The ability to enhance sensitivity and improve the accuracy and precision of the measurement translates into less material needed for analysis, decreased analysis time, and it opens the door for other analytes of interest not detectable by current methods.

Data was obtained by combining Vacuum Ultraviolet (VUV) laser postionization with dynamic SIMS. SIMS is a mass spectrometric method that generates secondary ions (positive and negative) by impacting solid surfaces with high-energy primary ions (Cs⁺, O⁻, etc.). For the SIMS instrument at Pacific Northwest National Laboratory (PNNL), the secondary ions represent approximately one percent of all material removed during the analysis with the rest remaining neutral. The aim of postionization is to photoionize a large fraction of the remaining neutral material using a high-energy, focused laser and achieve ionization efficiencies approaching 100%. Development of the VUV optical setup and modifications to the existing IMS-4F SIMS instrument at PNNL to accommodate laser postionization will briefly be discussed.

Preliminary data was collected using pure metals (In, U, Bi, and Re) in order to demonstrate proof-of-concept. Ions can be generated in several ways during these experiments; however, postionization signal enhancements depend on proper overlap of both the primary ion beam and the photoionization laser. Time-resolved experiments were performed with the primary ion beam (on/off) and laser (on/off) to confirm the observation of postionization signals. The metal standards were specifically chosen because they represent elements with ionization potentials above (In and U) and below (Bi and Re) the photon energy (6.4 eV) of the VUV laser. The original hypothesis was that the postionization signals for In and U would be significantly higher than for Bi and Re given the photon energy of the laser. Results contradict that hypothesis and demonstrate that relatively equivalent postionization signals were measured for all the chosen metals. This observation indicates that neutrals are desorbed into hyperthermal energy states. Current and future work is focused on using continuous wave lasers versus the pulsed laser used for this initial study, which is expected to substantially increase the duty cycle for these types of experiments.

Although in its infancy, the research presented herein has great potential to increase the sensitivity for mass spectrometric analyses of nuclear materials and directly support nuclear non-proliferation and other national security missions.

Mass Spectrometry, VUV Laser, Increased Sensitivity

A29 Coherent Anti-Stokes Raman Scattering Microspectroscopy of Fingerprints

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After attending this presentation, attendees will gain an understanding about the importance and usefulness of the methylene group in detecting and imaging untreated fingerprints by vibrational spectroscopy.

This presentation will impact the forensic science community by demonstrating that Coherent Anti-Stokes Raman Scattering (CARS) provides a new technique advantageous for reducing background noise in imaging fingerprints.

Vibrational spectroscopy, such as mid-Infrared Reflection (IR) or absorption spectroscopy and Raman Scattering Spectroscopy (RS), has been considered as a powerful tool to detect untreated latent fingerprints.¹⁻⁵ Generally speaking, each has its own drawbacks. The spatial resolution is poorer in IR than in RS. As Raman signals are usually dominated by Stokes signals, Raman signals are often hidden by fluorescent spectra. CARS has sufficient spatial resolution on the same level as RS and its signal is free from fluorescence though it comes with a non-resonant contribution. Thus, CARS is considered to be one of the promising techniques to overcome the drawbacks. In this report, CARS signals and images of untreated fingerprints on various backgrounds are presented.

CARS microscopic system was configured with commercially available instruments. A mode-locked laser was used as a laser source. Typical duration, peak wavelength, and repetition rate were 100fs, 800nm, and 80MHz, respectively. A portion of the output from the oscillator was used for a seed laser to generate a supercontinuum in the fiber module. The fundamental of oscillator and the supercontinuum were used as pump and Stokes lasers, focused onto a sample on an XYZ automatic stage with a 20×0.4 NA microscopic objective. CARS signals were dispersed by imaging spectrograph, and detected by an Electron Multiplying Charge Coupled Device (EMCCD) camera. The spatial and spectral resolution were 2μm and 0.94nm, respectively. Untreated fingerprints on a glass slide, a copper sheet, and white photo paper were prepared as samples. As necessary, either forward or backward scattering configuration was used.

Just after fingerprinting, CARS signals were obtained successfully from the fingerprint on the slide in both the forward and backward scattering configurations. Decomposition procedures of the signals revealed that the resonant Raman signal appeared at 2850cm⁻¹.⁶ Its origin was attributed to the C-H stretching vibrational mode of the methylene group. CARS images were constructed using signals averaged over a strong band around 2850cm⁻¹. In the forward scattering configuration, clear images were obtained for the fingerprints on the slide. Similarly, clear images were obtained for the fingerprints on the copper sheet and white photo paper in the backward scattering configuration. Temporal changes in the CARS signals of the fingerprints were also studied. The outline of the fingerprint was still imaged for the latent fingerprint on slide 30 days later. A clear image was obtained for the fingerprint on white photo paper several days later. The latter is a remarkable finding because it is known that any previous methods are not capable of clearly imaging such a latent fingerprint on white photo paper.

In conclusion, the present results suggest that there are cases when the CARS technique and the previous ones are complimentary each other.

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CARS, Untreated Fingerprints, Vibrational Mode of Methylene

A30 Biological Evidence Preservation: Guidance for Evidence Handlers

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After attending this presentation, attendees will become aware of the recommendations of the recently published report developed by NIST and NIJ entitled, *The Handbook on Biological Evidence Preservation: Best Practices for Evidence Handlers*.

This presentation will impact the forensic science community by bringing awareness to issues in biological evidence storage and the solutions proposed by a working group of experts in forensic science, law enforcement, property and evidence storage, and law.

The proper long-term storage and preservation of biological evidence has become increasingly newsworthy as states throughout the U.S. enact legislation allowing post-conviction DNA testing of evidence. Recent headlines have highlighted significant problems with the storage of potentially exculpatory biological evidence in property and evidence storage units across the country. To address these issues, the National Institute of Justice (NIJ) has provided funding for the National Institute of Standards and Technology's (NIST) Law Enforcement Standards Office (OLEs) to lead a technical working group charged with examining current policies, procedures, and practices in biological evidence storage. The primary objective of the working group is to establish science-based best practices to reduce the premature destruction and degradation of biological evidence, thus ensuring its availability for future analysis.

The group's analyses of the current state of biological evidence preservation in property and evidence rooms have shown that there is significant fragmentation and a lack of standards, particularly for long-term storage of biological evidence. Court orders for the location of evidence have demonstrated inadequacies in the packaging, storage, and tracking process of some evidence. Investigations into these inadequacies have revealed underlying factors such as: capacity of the storage facility, laboratory backlog, materials available for packaging, geographic distance between the collecting and storage facilities, and, the selected tracking system. Further complicating this issue, a number of states have enacted post-conviction DNA testing, resulting in more evidence being stored. Conversely, inquiries into exonerations have been delayed or thwarted in some cases due to problems already in existence in the property room. The disposition of evidence is a key consideration as the majority of evidence stored is of questionable probative value. Management of evidence in many long-term storage locations is lacking. Accordingly, jurisdictions must develop standardized protocols to identify what evidence to keep, how to keep it, and for how long.

The group's key deliverables can be found at <http://nist.gov/oles/forensics/bioev.cfm> and include a handbook outlining best practices and standardized protocols for property and evidence clerks, a report on legislative considerations, a report discussing current technological trends and possible applications, and a web-based clearinghouse for biological evidence handlers in property rooms, courts, and law enforcement agencies.

Evidence, Biological, NIST

A31 Detecting Spermatozoa Using the Proximity Ligation Real-Time PCR (PLIRT-PCR) Method

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After attending this presentation, attendees will be informed as to how the PLIRT-PCR method can be used to detect spermatozoa for forensic applications.

This presentation will impact the forensic science community by demonstrating that using the PLIRT-PCR method to confirm the presence of spermatozoa can potentially replace the current, time-consuming, microscopic detection method. The PLIRT-PCR method is a fast, sensitive, specific, low consumption, high-throughput, and amenable-to-automation molecular assay that could aid in reducing the backlog of processing sexual assault evidence.

In sexual assault cases, the confirmatory identification of spermatozoa often plays a critical role in determining whether or not a sexual act occurred. In some crime labs, the detection of semen begins with the seminal acid phosphatase presumptive test followed by the significantly more specific Prostate Specific Antigen (PSA or p30) test. The latter cannot be considered confirmatory for semen because PSA has been detected in other body fluids such as male and female urine and breast milk.

The only universally accepted method to confirm the presence of semen today is to visualize spermatozoa microscopically. This is a time-consuming task, as each sample must be processed and examined individually. Due to the large number of sexual assault cases that are processed in forensic laboratories, the detection of semen significantly affects case backlog. Automated fluorescence-based microscopic detection can decrease the time spent per sample, but still processes one sample at a time and requires expensive equipment.

Presence of semen can also be confirmed through the detection of sperm cell specific proteins. The PLIRT-PCR assay is designed for the detection of proteins, combining the specificity of an immunological reaction with the sensitivity of PCR. The process involves creating a DNA representation of a particular protein upon the dual binding of specific antibody assay probes. DNA surrogates of proteins are created by proximity ligation and detected via real-time PCR. The amount of signal, identified by the cycle threshold (Ct), is inversely proportional to the original amount of amplicon, which in turn indicates the amount of protein in the sample. The PLIRT-PCR technique only requires a thermal cycler and a real-time PCR instrument, both commonly used in forensic labs.

A PLIRT-PCR assay was developed using the TaqMan[®] Protein Assay Kits by Life Technologies[™] to target the sperm-specific protein SP-10 (ACRV-1). SP-10 is an intra-acrosomal, testis-specific protein that is an ideal target for the assay because

the acrosomal location protects the protein from environmental elements and its location on the surface of the acrosome makes it likely that epitopes will be accessible to the antibody-probes.

Preliminary work has shown that the PLiRT-PCR assay has been sensitive enough to detect SP-10 in liquid semen dilutions down to 1:5000, equivalent, in the assay, to about 10 spermatozoa. Each dilution from 1:10 to 1:10,000 was run in duplicate along with a No Protein Control (NPC). Ct values of semen dilutions outside of 3 standard deviations from the average NPC value are considered positive. A specificity test has also shown the assay to be specific only for semen samples when tested against both male and female blood, saliva, and urine. In the specificity testing, each body fluid sample was run in duplicate as well as with an NPC. The assay has also been evaluated for its ability to detect semen in common forensic samples such as swabs with mixtures of blood and semen and mixtures of saliva and semen. The samples tested consisted of a buccal swab with 50 microliters of a 1:10 dilution of semen added, a buccal swab alone, a swab with 50 microliters of whole blood with 50 microliters of a 1:10 dilution of semen, and a swab with 50 microliters of whole blood alone. Samples were run in triplicate along with an NPC swab. Only the samples with semen added were positive.

Although these early results are promising, some issues have been encountered with reproducibility and variability. For example, in certain experimental sessions, there was unexpected variability between Ct values obtained from aliquots of the same semen dilution and those tested in triplicate. These issues will continue to be resolved as the assay development continues. This presentation will discuss the experiments conducted thus far using the PLiRT-PCR method targeting SP-10, the numerous benefits of using a molecular method to detect spermatozoa, and the potential for targeting other or multiple sperm specific proteins with this assay.

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Spermatozoa, PLiRT PCR, Sexual Assault

A32 Identification of Marker Proteins in Menstrual Blood Using LC-MALDI Mass Spectrometry

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After attending this presentation, attendees will understand some past and present techniques for identifying menstrual blood, advantages to using proteomics for the analysis of menstrual blood, and some specific factors that need to be considered when identifying marker proteins in menstrual blood.

This presentation will impact the forensic science community by presenting a new serological method for identifying body fluids and provide a confirmatory test for menstrual blood.

Body fluid identification is an important tool to help investigators reconstruct details of a crime. While the more common body fluids (blood, semen, and saliva) have well-documented presumptive testing procedures, the lesser seen body fluids (vaginal fluid and menstrual blood) do not. Menstrual blood consists of blood, vaginal fluid, and endometrial cells. Due to this complex mixture of other known body fluids, current serological tests are not ideal for identifying menstrual blood because they

look for components of red blood cells, such as hemoglobin, which are also located in venous blood. Any new technique developed for the purpose of identifying menstrual blood will need to identify a substance unique to menstrual blood in order to be effective. Several techniques under development aim to distinguish menstrual blood from venous blood, including mRNA analysis, microRNA analysis, DNA methylation analysis, Raman spectroscopy, and protein analysis by mass spectrometry.

In protein analysis, proteins are digested into peptides and the masses of peptides are determined by tandem mass spectrometry. These data are compared to protein libraries and peptide/protein identifications are made. In recent years, most of the commonly occurring forensic body fluids have had their proteomes mapped using mass spectrometry. Recently, a laboratory has published the first menstrual blood proteome.¹ By comparing the menstrual blood proteome with other proteomes, it is possible to identify a group of proteins unique to or enriched in menstrual blood. Once a list of these proteins has been developed, the identity of any unknown sample analyzed on a mass spectrometer can be determined. Previous studies have shown proteomics to be highly sensitive and specific in venous blood, semen, and saliva identification.²

Samples of menstrual blood were collected from 45 women on each day of her period using a menstrual cup. Menstrual blood samples were homogenized and proteins were extracted using differential centrifugation. Protein concentration was measured using a bicinchoninic assay. Samples were processed using a protein enrichment kit to reduce the dynamic protein range and improve marker detection on the mass spectrometer. Samples were qualitatively analyzed by electrophoresis to evaluate the enrichment. Protein was digested using trypsin and labeled using isobaric tags to allow all samples from a single individual to be run on the mass spectrometer at the same time. Samples were separated using High Performance Liquid Chromatography (HPLC) and analyzed Matrix-Assisted Laser Desorption/Ionization-Tandem Time Of Flight (MALDI-TOF/TOF). The resulting spectra were compared to a protein database and any unique or highly enriched proteins determined. Results of the mass spectrometer and protein database comparisons and any candidate proteins will be discussed.

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Serology, Menstrual Blood, Proteomics

A33 Optimized Methods for Isolation of microRNAs From Forensically Relevant Body Fluids

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After attending this presentation, attendees will understand the basic biogenesis and function of microRNAs (miRNA), as well as how miRNAs can advance serological testing over traditional methods. The presentation will discuss which miRNA isolation methods were found to be most effective for each forensically relevant body fluid, and variations found in common miRNAs levels, not only between individuals in the same body fluid, but also between different body fluids from the same person.

This presentation will impact the forensic science community by introducing the community to a new potential molecular technique for body fluid identification. This application would permit a more concrete identification of body fluids in forensic cases, allowing for more accuracy in casework and confidence during testimony.

Current serological techniques are often subjective and typically do not have a statistical measure of confidence associated with the results. Because of this, the popularity and confidence level in DNA profiling has far outpaced the technology of body fluid identification; however, the accurate identification of body fluids present at crime scenes has not diminished in importance. While each body fluid has a variety of current and historical presumptive tests for forensic detection, almost all lack a specificity and sensitivity that would give examiners full confidence when identifying those body fluids. Molecular-based serological techniques are able to provide a confidence level that is currently lacking in traditional methods.

miRNAs are small non-coding RNA molecules that regulate many cellular processes through modulation of proteins at the translational level. miRNAs are very stable, both because of their small size and also due to the fact that when they are secreted, they are typically either encased in a protective lipid vesicle or encapsulated in a protein or cholesterol matrix. These properties make miRNAs more resistant to degradation than naked nucleic acids. Thus, it is likely that miRNAs, when fully evaluated, will make excellent candidates for body fluid identification. Identifying miRNAs that are specific to a particular body fluid could give an examiner absolute confidence when identifying that body fluid, an advantage that serological testing cannot provide.

In order to determine the most optimized isolation method that yields the highest quantity of miRNA for each body fluid, five commercial miRNA isolation kits were tested on blood, semen, menstrual blood, vaginal fluid, urine, sweat, and saliva samples. The five methods were first compared on efficiency of miRNA extraction by total RNA yield through UV spectroscopy, with the top three most efficient kits for each body fluid selected for further analysis. Vaginal fluid and menstrual blood had the highest average yield of total RNA with over 3,000ng and 2,600ng respectively, while urine and sweat had the lowest average yield, containing only 325ng and 250ng of total RNA respectively.

The extracted samples were evaluated using Real-Time Quantitative Polymerase Chain Reaction (RT-qPCR with known endogenous miRNAs and small RNAs in order to evaluate relative miRNA levels for each body fluid. miRNA levels were evaluated based on relative abundance, with samples yielding highest Ct values interpreted as having the highest quality and quantity miRNAs. Of the five kits, either the Qiagen® miRNeasy Mini Kit (menstrual blood, semen, saliva) or the Qiagen® miRNeasy Serum/Plasma Kit (blood, vaginal fluid, urine, sweat) was chosen based not only on effectiveness of extraction but also on ease-of-use.

Additional comparisons were made to evaluate the consistency of miRNA abundance levels. To determine the relative

abundance level of known endogenous miRNAs and small RNAs among individuals, a comparison of four individuals was made for each body fluid. This data showed that of the two miRNAs assayed blood, semen, and sweat levels varied very little (<3 Cts, or 8-fold) regardless of contributor, but urine, menstrual blood, and vaginal fluid had widely varying levels of the same miRNA (4 – 7 Cts, or up to a 128-fold difference).

miRNA isolation could increase the accuracy and efficiency of identifying body fluids by taking serological testing to a molecular level. The first step in this process is to optimize the most efficient isolation method to use for each body fluid to ensure the most accurate results.

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Forensic Science, Body Fluid Identification, microRNA

A34 Epigenetic Methylation Markers as a Powerful Technique to Discriminate Body Fluids Present in Crime Scenes

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After attending this presentation, attendees will gain valuable insight on the research efforts being made to use patterns of DNA methylation at different genome locations as a tool to discriminate body fluids present at crime scenes.

This presentation will impact the forensic science community by showing that epigenetic differences in DNA permit discrimination of cell type. A method has been developed to exploit the relative levels of methylation at CpG islands on the genome using bisulfite modification and pyrosequencing. Using this knowledge, evidence will be provided on how different body fluids present at crime scenes show distinctive methylation patterns at specific loci.

In certain criminal cases such as sexual assaults, discrimination between the type of cells present can assist the trier-of-fact in determining if a crime was committed or if the presence of suspect DNA derives from innocent contact. In general, current methods used to determine the presence of body fluids are presumptive and lack specificity and sensitivity. Alternative tests based on gene expression using mRNA markers have been developed (as have proteomic markers); however, these methods ultimately are derived from RNA.¹⁻⁶ Thus, use of epigenetic tests would be more direct and should be more stable with time.

The epigenome varies according to gene expression and ultimately permits cell differentiation within the human body. Eckhardt *et al.* described specific regions where DNA is either methylated or not according to the type of body tissue present, making them excellent markers for body fluid determination.⁷ The greater stability of DNA when compared to RNA and protein makes it the most suitable biomolecule for use in forensic discrimination of body fluids present in crime scenes.

In this study, DNA obtained from samples of semen, saliva, blood, skin, as well as old stains from blood stored on FTA® paper and in other materials were examined. DNA samples from non-humans such as *E. coli*, dog and cat were also tested to confirm species-specificity for the markers. All DNA was bisulfite modified using the Epitect® Bisulfite Kit to convert all the non-methylated cytosine's to uracils on the template DNA. Bisulfite modified DNA was then singleplex amplified using the primers designed by the PyroMark® CpG Assays that are gene-specific methylation assays for pyrosequencing analysis. All pyrosequencing reactions were performed using the PyroMark® Q24 Pyrosequencer. Data analysis was performed using the PyroMark® Q24 assay software for CpG methylation quantitation. The corresponding percent (%) methylation values for each site and the data were displayed as a pyrogram.

The data obtained shows successful discrimination of body fluids when specific markers are screened for methylation patterns, not just in DNA extracted from fresh samples but also in samples stored for several years. The C20orf117 marker discriminates blood with methylation levels between 50% and 82% for blood and below 30% for other body fluids. The BCAS4 marker can reliably identify saliva since it produces methylation levels 25% and above while other fluids tested show levels below 20%. For the ZC3H12D locus, all body fluids except semen/sperm show methylation levels above 80% whereas that number is below 13% for sperm. Also available to differentiate sperm, the marker FGF7 shows 60% methylation or greater for all 7 CpG islands in the locus while all other cell types are 30% or below. Non-human DNA is not successfully sequenced using any of the selected primers.

In total, the results show a differential methylation pattern for specific loci that can be used to discriminate diverse body fluids. This method is more specific than the currently used protein-based techniques because it does not exhibit cross-reactivity between body fluids. Further, this method is robust, permitting use on samples stored in evidentiary archives.

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Epigenetics, DNA Methylation, Pyrosequencing

A35 Development and Testing of a Mass Spectrometry-Based Assay for the Identification of Biological Stains

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After attending this presentation, attendees will be aware of a novel method for the classification of biological stains through the use of targeted mass spectrometry.

This presentation will impact the forensic science community by informing it of a novel technology with the potential to significantly improve the accuracy and sensitivity of forensic serological testing. It will provide practitioners with greatly improved tests for saliva, seminal fluid, peripheral blood, and urine while also enabling the identification of vaginal and menstrual fluids for which there are currently no accurate tests.

While DNA profiling makes it possible to individualize biological stains, the identification of the stain itself can present forensic serologists with a significant challenge. For example, if a genetic profile generated from a swab of a male suspect's finger is consistent with that of an alleged female victim of sexual assault, more than one interpretation is possible. Did the male sexually assault the victim by digitally penetrating her vaginally? Or, did the female lick the suspect's finger — leaving her DNA behind? The ability to reliably determine whether the suspect's finger contains traces of saliva and/or vaginal secretions could enable investigators to better evaluate such alternate interpretations.

Current antibody and enzyme activity-based assays used by forensic practitioners for biological stain testing yield only presumptive results. Positive results with non-target body fluids or cross-reactivity with non-human sources has been well documented. Some serological tests can consume unacceptable quantities of precious evidence, while for some body fluids there are simply no available tests at all. There is clear value in developing more accurate approaches for identifying stains of biological origin.

Using state-of-the-art protein characterization technologies, the proteomes for six body fluids (peripheral and menstrual blood, vaginal fluid, semen, urine, and saliva) have been mapped and characterized. Based on these analyses, 37 protein biomarkers were identified for the confirmatory identification of all target stains. These candidates included several well-known markers, such as Statherin for saliva identification, as well as many novel biomarkers such as matrix metalloproteinase-9 for vaginal fluid identification.

Verification assays have been performed to confirm the specificity and consistency of the most promising candidate biomarkers across a large human sample population. Verification assays employed a targeted, multiplex, mass spectrometry assay on a nanoflow quadrupole time-of-flight instrument. Results of these studies have identified a panel of accurate, reliable, and confirmatory protein markers for biological stain identification. The ability to detect the markers in a variety of casework-type samples was also successfully tested.

Serology, Mass Spectrometry, Proteomics

A36 Separation of Complex Biological Mixtures Using Human Leukocyte Antigen Probes and Fluorescence Activated Cell Sorting (FACS)

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After attending this presentation, attendees will gain an understanding of how molecular probes targeting antigens on white blood cells can be used to differentiate individual contributions to a complex blood mixture and the application of Fluorescence Activated Cell Sorting (FACS) for physically separating cells prior to Short Tandem Repeat (STR) analysis.

This presentation will impact the forensic science community by introducing a new technique to resolve individual contributions in a biological mixture. Molecular antibody probes coupled to flow cytometry has the potential to reduce analytical bottlenecks within case working laboratories and the loss of evidence that can accompany the interpretation of STR profile mixtures.

Analysis of biological mixtures is a significant problem for forensic laboratories. The presence of cells from multiple individuals in a biologic stain complicates DNA profile interpretation and often leads to loss of evidence. While many analytical techniques have been developed to address complex STR profiles resulting from cell mixtures, few can be applied to biological mixtures containing one cell type. Yet, an increasing proportion of samples submitted as evidence are mixtures in which there are no physical differences between cells contributed by multiple individuals. New techniques are needed to separate individual cell populations from these types of forensic mixtures to generate unambiguous STR profiles.

To address this problem, a new analytical technique was developed that utilizes the intrinsic immunological variation among individuals to physically separate cells from different sources prior to DNA profiling. For this procedure, fluorescent antibody probes are used to selectively bind to allelic variants of the Human Leukocyte Antigen (HLA) complex on cell surfaces. The natural diversity of antigen alleles and their universal expression on the cell surfaces make them ideal molecular targets for differentiating contributors in a mixture. Once individual cell populations are labeled with HLA-antibody probes, they are isolated from the mixture through FACS, a high-throughput technique for separating cell populations based on their optical properties. By separating whole cells from a forensic mixture prior to genetic analysis, single-source STR profiles could potentially be generated without any ambiguity or complex pattern interpretation.

The forensic potential of this technique was tested on a two-person whole blood mixture. A discriminatory antigen target (HLA-A02) was identified prior to the experiment and the complementary antibody probe was used for all separation procedures. Specificity of the HLA-A02 probe to the surface antigens of each contributor was checked prior to mixing samples. First, one microliter of whole blood from each contributor was mixed together and the liquid mixture was hybridized with the HLA-A02 fluorescent probe. Characterization with flow cytometry showed two cell populations, each with distinct fluorescence profiles. Optical characteristics of each cell population were consistent with antibody probe binding with only Contributor 1 cells in the mixture. Isolation of the labeled cell population was conducted by FACS. DNA extraction and STR profiling of the labeled cell population was performed to determine the efficiency of cell separation. Profiles were compared against STR profiles generated for the whole blood mixture and reference samples from each contributor. Results showed that the STR profile of the fluorescently-labeled cell population that was sorted from the mixture were identical to the single source profile for Contributor 1 across all six Combined DNA Index System (CODIS) loci tested. Further, there was no evidence of allelic contributions from Contributor 2 after cell sorting. Antibody labeling and FACS sorting were also tested on liquid blood mixtures containing different ratios of cells from each source (1:2, 2:1, 3:1, 1:3). Resolvable single-source STR profiles were obtained for 1:2 and 2:1 mixtures; however, sorted cell populations from 1:3 and 3:1 mixtures yielded no interpretable allelic peaks. This indicates that the individual template ratio has an effect on the efficacy of antigen

labeling and cell sorting and the procedure needs to be optimized for these types of mixtures.

Overall, these experimental results suggest that HLA-antibody probes can be used to differentiate individual contributors in a complex mixture and that FACS is an efficient way to physically separate antibody-labeled cells for single-source STR profiling. This technique has the potential to help reduce the analytical bottlenecks, inconclusive results, and loss of evidence that often accompany mixed STR profile interpretation within forensic case working units.

STR Profiling, FACS, Antibody Probes

A37 The Role of Alkaline Lysis and Pressure Cycling Technology in DNA Recovery From Mixtures

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After attending this presentation, attendees will understand the principles behind a rapid and efficient method to extract sexual assault evidence using pressure-based protocol and the effect of alkaline lysis on sample recovery from cotton swabs.

This presentation will impact the forensic science community by introducing the application of pressure pulsing for specific detection of individual cells in mixed stains which will enable rapid and selective processing of sexual assault evidence.

Sexual assault cases are plagued by challenges in sample analysis due to the presence of tissue from multiple contributors. The sample mixtures are often overwhelmed by female epithelial cells which makes generating a clean male DNA profile very difficult. To overcome this obstacle, organic differential extraction is most commonly used, wherein the overwhelming female fraction in the mixture is lysed and removed followed by the lysis of sperm cells. Apart from the tedious procedure and incomplete mixture resolution associated with differential extraction, the inability to efficiently recover the sample from the cotton matrix contributes to the challenges faced in analysis of mixtures.

The objective of this study was development of a method using Pressure Cycling Technology (PCT) combined with reagents to selectively disrupt sperm or epithelial cells and recover DNA. The extraction procedure was performed utilizing the Barocycler® NEP 2320, a commercially available instrument equipped with a hydrostatic pressure chamber that generates alternating cycles of ambient and high pressures with a range of 5-45kpsi. Another goal of this study was to enhance sample recovery from cotton swabs. Cotton swabs are often used to collect crime scene evidence; however, the inefficient sample recovery from this substrate has been the subject of numerous studies in the past. In order to enhance DNA recovery from cotton swabs and hence improve downstream genetic analysis, the effect of alkaline lysis on sample recovery from this substrate was studied.

The current study involved the application of pressure cycling technology in the selective digestion of sperm cells from evidence mixtures with an emphasis on the role of buffer composition on sperm DNA yields and increase in selectivity of extraction. The cells were extracted into 1X PBS buffer (pH 7.4) with varying buffer compositions and subjected to 45,000psi pressure. To improve sample recoveries, cotton swabs containing sample were incubated in different concentrations of sodium hydroxide under varying temperature and incubation times. Following incubation, all the samples were purified using phenol/chloroform/isoamyl alcohol extraction and quantified with the Promega® Plexor® HY System. The samples were subsequently typed by Short Tandem Repeat

(STR) analysis using the Promega® PowerPlex® 16 HS System.

Preliminary results indicated that the application of pressure cycling technology, in the presence of appropriate buffers, can result in 50-60% recovery of male DNA from mixtures. These observations were reproduced with mixtures on cotton swabs where six times more male DNA was recovered compared to female epithelial DNA. Furthermore, preliminary data from the alkaline lysis studies indicated significant increase in DNA recoveries from cotton swabs with an increase in selectivity for sperm DNA recovery. Careful optimization of parameters including NaOH concentration, incubation time, and temperature resulted in 5- to 6-fold enhancement in the relative ratio of sperm DNA in these mixtures.

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Pressure Cycling Technology, Alkaline Lysis, Mixtures

A38 Comparison of Reducing Agents in Differential DNA Extraction

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After attending this presentation, attendees will learn how Dithiothreitol (DTT), the most common reducing agent employed in differential extraction procedures, compares with other reducing agents in their ability to generate male DNA profiles from dried stains prepared from female saliva and seminal fluid.

This presentation will impact the forensic science community by demonstrating that the effectiveness of differential extraction procedures for the isolation and subsequent genotyping of DNA can be improved with the use of a new reducing agent, dithiobutylamine, or beta-mercaptoethanol.

Differential extraction procedures have been utilized on sexual assault evidence for two decades and involve the selective digestion of epithelial cells followed by isolation and digestion of sperm cells. A reducing agent is used to lyse sperm so that DNA from these cells can be isolated. Although several alternatives are available commercially that have increased throughput, many laboratories still use the traditional Chelex® or organic differential extraction method due to problems related to cost and implementation. Recently, a new reducing agent, Dithiobutylamine (DTBA), was synthesized from L-aspartic acid and found to be far superior in reducing disulfide bonds in aqueous solution when compared to several reducing agents, including DTT.¹ Reduction of disulfide bonds on the membrane of sperm cells is necessary to isolate sperm DNA in these procedures. As a result of this research, a pilot study was conducted to see if male DNA profile quality could be improved by comparing the use of DTBA in a differential extraction procedure, not only to DTT but to other reducing agents as well.

Simulated post-coital swabs were created by adding 2 μ L of neat semen, 1:10 diluted semen, and 1:100 diluted semen, respectively, to buccal swabs and allowing them to dry. Differential Chelex® extraction was performed in triplicate on each set of swabs using DTT, DTBA, beta-mercaptoethanol, or tris(2-carboxyethyl) phosphine, respectively.

A one molar (1M) concentration of each reducing agent tested was used with a volume of 7 μ L added during extraction. Extracted DNA was quantified using the Alu-based real-time

PCR SYBR green method and appropriate DNA concentrations were amplified using the Promega® PowerPlex® 16 HS System. Fragment analysis of amplified products was performed on an ABI Prism 310 Genetic Analyzer and amplicons were genotyped using GeneMapper[®] ID-X Software.

Results from swabs containing neat semen showed no difference in the ability to obtain a sperm DNA profile in differentially extracted samples using DTT, DTBA, or beta-mercaptoethanol. Triplicate swabs using these reducing agents all produced full sperm profiles. No male DNA was detected when tris(2-carboxyethyl) phosphine was used as the reducing agent. As a result, none of the swabs which used tris(2-carboxyethyl)phosphine as a reducing agent were genotyped.

The quality of sperm DNA profiles did vary with swabs containing 1:10 and 1:100 diluted semen with DTT, DTBA, and beta-mercaptoethanol. DTT did not produce any full profiles with swabs containing either 1:10 or 1:100 diluted semen. When using DTT as the reducing agent, the male fraction averaged 22 and 12 alleles (out of a possible 26) for the 1:10 and 1:100 diluted semen swabs, respectively.

Both DTBA- and beta-mercaptoethanol-treated swabs produced more complete sperm DNA profiles than DTT. With DTBA, an average of 25 male alleles (out of a possible 26) was detected on the 1:10 diluted semen swabs and two of the three swabs produced full male profiles. With the 1:100 diluted semen samples, DTBA treated samples averaged 16 male alleles (out of a possible 26) and no full profiles were obtained. All three swabs containing 1:10 diluted semen produced full profiles with beta-mercaptoethanol and an average of 17 male alleles (out of a possible 26) were detected on the 1:100 diluted samples. No full male profiles were obtained on the swabs containing 1:100 diluted semen when beta-mercaptoethanol was used as the reducing agent.

Results from this pilot study indicate that both DTBA and beta-mercaptoethanol used as a reducing agent in differential extraction may be superior in developing sperm DNA profiles from male-female mixed samples than the commonly used DTT, particularly with samples containing a small volume of semen.

Reference:

1. Lukesh JC, Palte MJ, Raines RJ. *J Am Chem Soc* 2012;134:4057-9.

Differential Extraction, DTBA, Beta-Mercaptoethanol

A39 Elemental and DNA Analysis of Constituents Obtained From Extracting Bone Samples

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This presentation will illustrate the separation of sample constituents as a consequence of DNA extraction from human skeletal remains. Presented data include elemental contents, DNA quality and quantity, and genetic analyses. Results were obtained by examining working and waste fractions from each step of a DNA extraction process, a modification of the Loreille

et al. demineralization method with an additional silica-based purification.¹ After attending this presentation, attendees will have an understanding of how nucleic acids and elemental materials are lost or carried over through each step of this extraction process.

This work will impact the forensic science community by attempting to increase the number of individuals identified from skeletal remains by improving the development of techniques for extracting and analyzing DNA from bone samples. These are considered one of the most challenging sample types in the laboratory and require laborious processing that frequently yields incomplete genetic results or amplification failure. Possible causes of poor results may include DNA damage, PCR inhibitors, and/or inefficient extraction. These experiments examined the separation of materials during DNA extraction to measure the efficiency of current methods. Understanding the complexity of human bone samples and the efficacy of each step in routine laboratory processing will provide a foundation for the future development of improved methods to extract DNA from skeletal remains.

Pulverized bone samples from adjudicated cases were obtained from the University of North Texas (UNT) Center for Human Identification. Separate, homogenized aliquots of each bone sample were used for chemical and DNA testing. Baseline elemental content was determined by Inductively Coupled Plasma-Mass Spectrometry (ICP/MS). Elemental analysis was performed in solution mode ICP/MS. Acid digestion was used for sample preparation; approximately 100mg of each sample was dissolved in ultra-pure concentrated nitric acid (HNO₃) and then diluted with 1% HNO₃. For major and trace metal analysis, dilution factors varied between 20 and 100. All samples were measured in triplicate for statistical analyses.

DNA extraction was performed on the second aliquot of each bone sample using the UNT Center for Human Identification "Demineralization Extraction of Skeletal Remains" protocol. This procedure is a modified version of the total demineralization extraction by Loreille *et al.*¹ Samples were weighed and placed into polypropylene test tubes. Demineralization buffer and proteinase K were added to each sample and reagent blank and incubated overnight. An equal volume of Phenol/Chloroform/Isoamyl Alcohol (PCIA) was added to each sample, vortexed and centrifuged. Aliquots for subsequent testing were removed from the aqueous phase. While the waste fraction from the PCIA step could not be tested due to the material hazards present, the remaining aqueous phase was transferred to a centrifugal filtration device. Samples were centrifuged through the device and aliquots were taken from the filtrate that had passed through the device and the retentate. The remainder of the retentate was additionally purified using a chaotropic salt solution and a silica-containing centrifugation column. Aliquots were also taken from the filtrate and retentate from this step for additional testing.

A total of five fractions were examined: the aqueous phase following PCIA purification, the filtrate and retentate from the centrifugal filtration, and the filtrate and retentate from the additional purification. Analyses targeting DNA content were conducted, including: DNA sizing and quantity on a microfluidics-based platform; UV-Vis spectrophotometry, PCR-based DNA quantification; STR amplification; and, genetic analysis on a capillary electrophoresis instrument. Elemental analysis of aliquots from each fraction was performed using ICP/MS.

No sizing results were produced for many sample sets using the microfluidics-based platform, believed to be a consequence of the reagents present in those fractions; however, gel images were obtained for each fraction tested. The results of the UV-Vis spectrophotometric analyses demonstrated an increase in A260:A280 ratio that corresponded with a significant decrease in estimated quantity of nucleic acids through each step of the extraction process. PCR-based DNA quantification produced some results and indicated the presence of PCR inhibitors in many

sample fractions. Results of the STR amplification and subsequent genetic analysis will be presented along with the elemental composition obtained from each fraction.

Reference:

1. Loreille OM, Diegoli TM, Irwin JA, Coble MD, Parsons TJ. High efficiency DNA extraction from bone by total demineralization. *Forensic Sci Int Genet.* 2007 Jun;1(2):191-5.

Skeletal Remains, DNA Extraction, Elemental Analysis

A40 Maximizing mtDNA Testing Potential With the Generation of High-Quality mtGenome Reference Data

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After attending this presentation, attendees will be informed regarding the development of more than 550 high-quality mitochondrial genome haplotypes and the successful completion of a project funded by the National Institute of Justice.

This presentation will impact the forensic science community by discussing soon-to-be available complete mitochondrial genome reference data which meet the highest forensic standards and a web-based search engine to permit their use for forensic casework.

Mitochondrial DNA (mtDNA) testing in the forensic context requires appropriate, high-quality population databases for estimating the rarity of questioned haplotypes to, in turn, determine the strength of the mtDNA evidence. However, at present, all publicly available forensic mtDNA reference databases Scientific Working Group on DNA Analysis Methods (SWGDM) and EDNAP mtDNA Population Database (EMPOP) only include information from the Control Region (CR). Emerging technologies such as next-generation sequencing are capable of producing complete mitochondrial genome (mtGenome) data from extremely low DNA quality and quantity forensic specimens, but no suitable public database of complete mtGenomes is available for forensic queries. Thus, the goals of this National Institute of Justice (NIJ) funded project were to: (1) increase the large-scale availability of high-quality entire mtGenome reference population data; and, (2) improve the information technology infrastructure required to access/search mtGenome data and employ that data in forensic casework. The specific objectives of this large-scale databasing initiative were the development of 550 high-quality mtGenomes spanning three U.S. population groups, along with database structure and query modifications to the publicly-available EMPOP database.

To ensure the generation of the highest quality mtGenome profiles, a laboratory processing workflow was implemented in which nearly all pipetting steps were performed robotically and a rigorous data review process was employed. Amplification of the complete mtGenome was achieved via eight overlapping fragments, and each mtGenome was sequenced in 135 reactions, providing redundant and overlapping forward and reverse sequence coverage across the entire molecule. The data review process followed a strategy previously used successfully for the production of high-quality CR sequences, which includes raw data review by no fewer than three distinct scientists at two laboratories and electronic data transfer with two additional profile reviews.¹ To further ensure data reliability, completed mtGenome haplotypes were compared to PhyloTree to confirm phylogenetic consistency across the eight amplicons.² Lastly, final profile haplogroups were assigned using an automated, maximum likelihood-based tool, EMMA.³

The presentation will describe the development of more than 550 entire mtGenomes using this workflow and the completion of the NIJ-funded databasing effort. The optimized, highly automated protocol reliably produced high-quality data from low template specimens and reduced the number of manual production steps and the extent of sample reprocessing necessary to construct complete mtGenome haplotypes. The efficacy of automated processing combined with a rigorous data review strategy in preventing errors was evident from the absence of problems detected at the stage of phylogenetic data evaluation. Few discordant profiles were identified upon cross-validation of the Armed Forces DNA Identification Laboratory (AFDIL) and EMPOP reviews.

In the immediate term, these high-quality haplotypes will be used as an etalon dataset for a posteriori quality control of mtGenome data evaluated by EMPOP prior to their acceptance for publication in two forensic journals. Ultimately, this project will provide the forensic community with reliable, complete mtGenome reference data and a means to access, search, and use the data in forensic casework.

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1. Irwin *et al.*, *Forensic Sci Intl: Genet* (2007) 1(2):154-7.
2. Van Oven and Kayser, *Hum Mutat* (2009) 30:E386-E394.
3. Röck *et al.*, *Forensic Sci Intl: Genet* (2013) <http://dx.doi.org/10.1016/j.fsigen.2013.07.005>.

Mitochondrial DNA, Population Database, Complete mtGenome

A41 Development of a Novel and Sensitive DNA Analysis Multiplex Based on INNUL Markers for Highly Degraded Forensic DNA Samples

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After attending this presentation, attendees will understand the development and application of a novel marker system for use with degraded forensic DNA samples.

This presentation will impact the forensic science community by serving as a key aspect for the analysis of degraded forensic DNA samples, as it can augment or replace traditional DNA testing systems in the cases where those systems do not produce results due to the presence of highly degraded DNA.

Retrotransposable Elements (REs) consisting of Long Interspersed Nuclear Elements (LINEs), Short Interspersed Nuclear Elements (SINEs), and SINE/VNTR/Alu (SVA) are a group of markers that can be useful for human identity testing. SINEs are a class of REs that are typically less than 500 nucleotides long; LINEs are typically greater than 500 nucleotides long. The third type of RE is the composite retrotransposon known as SVA elements.¹

One of the advantages of using REs is that they do not yield stutter artifacts due to slippage during the PCR. Until recently, however, due to the inherent size difference (>300bp) associated with insertion and null alleles, the use of REs has not been practical for forensic applications. LaRue *et al.* have termed the *Alus* and *LINEs* as *INNULs* instead of *INDELs*, as the two allelic states are known, to be an insertion and null; thus *INNUL* better describes the already well-established allelic states of these markers.² Although the use of SINEs such as *Alu* in human identity testing has been studied and reported in the literature, the more than 300bp size difference between the two alleles prevented development of any useful multiplexed system. This is the first time any research on the use of LINEs, SINEs, or SVA element insertions, as a multiplexed system, has been successfully achieved.

To circumvent the allele size disparity, a primer design methodology was used to remove the intra-specific locus competition that occurs in heterozygotes. This approach involved use of the direct repeat units that flank an *Alu* element. The novel primer design reduced overall amplicon size among loci as well as the difference in amplicon size between the two allelic states of *INNULs*. The resulting *INNUL* allelic amplicons can be designed to differ by as little as one base pair instead of the 300bp *Alu* insertion. Additionally, the amplicon size has been reduced substantially to a size smaller than currently used STR markers, such that substantially degraded DNA samples can be analyzed. Utilizing this primer design, a more simplified, rapid, and automated typing technology can be applied to LINE, SINE, and SVA insertion polymorphism typing.

Markers were selected based on population studies and analytical performance. The development of 16 REs and Amelogenin into a single multiplexed amplification system will be presented. The primer design was successful in substantially reducing the size of the amplicons. All 17 markers are between 50bp to 125bp. Example multiplex electropherograms showing the selected markers, fluorophores, and allele base pair sizes will be presented. The small amplicon sizes result in an extremely sensitive, rapid, and useful multiplex for highly degraded forensic

samples as well as for high-quality samples. When evaluating three major North American populations, these markers generally met Hardy-Weinberg expectations and showed little evidence of detectable levels of linkage disequilibrium between the markers tested, i.e., based on the exact test there were no more departures from expectations after Bonferroni correction. Random match probabilities were $<10^{-4}$. Known paternity trios were used to evaluate the system and demonstrate adherence to Mendelian inheritance principles.

Data from this study supports the usefulness of this system for analyzing highly degraded DNA samples which did not produce usable STR results, allowing results having high discrimination power to be obtained. A limitation of this system due to the bi-allelic nature is the inability to deconvolve mixtures by quantitation or by heterozygosity. This system will prove very useful for analyzing single source degraded DNA samples such as those found in mass disasters as well as other human identification efforts and, to some degree, will be applicable to mixture analyses.

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Alu, Degradation, INNUl

A42 Optimization and Evaluation of a Rapid PCR Method for a Commercially Available MiniSTR Kit for Human Identification

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After attending this presentation, attendees will have a better understanding of how using fast chemistry and rapid Polymerase Chain Reaction (PCR) methods can successfully produce Short Tandem Repeat (STR) profiles of acceptable quality for genetic analysis.

This presentation will impact the forensic science community by providing a new rapid PCR protocol for a miniSTR system that can increase sample throughput. The increased throughput will allow forensic laboratories to significantly reduce backlogs in cases involving low amount Deoxyribonucleic Acid (DNA) template or degraded samples that may have become more difficult to analyze.

Forensic DNA profiling is a four-step process consisting of DNA extraction, DNA quantification, PCR amplification, and separation and detection of the amplified DNA fragments via capillary electrophoresis. This process takes approximately 10h to 12h to complete, with PCR amplification requiring 3h to 4h. A reduction in the amount of time required for the amplification step would allow for faster human identification and increase laboratory throughput. This can be accomplished using fast chemistry and rapid PCR cycling instrumentation. Fast chemistry involves the application of new polymerases and buffers that maintain the levels of sensitivity and fidelity of standard polymerases but increase the extension rate and are activated more quickly. These fast chemistries are designed to be paired with rapid PCR cycling instrumentation where temperature transitions between annealing and extension PCR steps and holding times are shortened. The

goal of this work was to optimize and evaluate a rapid PCR method for the PowerPlex®S5 system for forensic DNA profiling.

Through the use of fast chemistries and a fast thermal cycler, the amplification time of the PowerPlex®S5 system was reduced. DNA profiles were produced in an hour, which was a 70% reduction from the standard protocol. Sensitivity and heterozygous peak height ratios were comparable between the fast and standard protocols. However, a substantial decrease (5%) in peak height ratio was noted at the D18S51 locus using the fast cycling method. Furthermore, an increase (2.6%) in the average mean stutter for combined loci was observed in profiles amplified using the fast protocol compared to the standard system. The results of this study demonstrate that a fast amplification protocol for a miniSTR system can be used to produce acceptable profiles for forensic DNA analysis. Further optimization and validation of the fast PowerPlex® S5 protocol are required before it can replace the standard amplification protocol in forensic laboratories.

Rapid PCR, Increased Throughput, Backlog Reduction

A43 Establishment of 11 Clustered X-STR Loci for Kinship Testing

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After attending this presentation, attendees will learn 11 new clustered X-chromosome Short Tandem Repeats (X-STR) loci for kinship test.

This presentation will impact the forensic science community by teaching a method of establishing new clustered X-STR loci for kinship test.

To increase the value of haplotype-linked X-STR loci in the kinship testing, reported is the identification and characterization of a cluster of 11 STR loci on the human X chromosome all within a range of 1.1 Mb. The repeat DNA sequences were identified, using data on GenBank, immediately upstream and downstream from the previously described locus DXS6807. Initially, there were 24 loci with four repeat motifs and three penta-repeats. The polymorphic nature of the loci was examined and only loci with four or more observed alleles were retained for further study. This resulted in the identification of ten new loci, on either side of DXS6807, and given the temporary names DXS680701, DXS680702, DXS680703, DXS680704, DXS680705, DXS6807, DXS680706, DXS680707, DXS680708, DXS680709, and DXS680710. The distance between each pair of loci ranged from 24,998bp to 244,701bp and average approximately 110.8kb. In this study of 141 unrelated female and 130 male samples, X-STR loci were successfully amplified by PCR and sequenced. From the resulting sequence alignments, the number of alleles at each of these loci was 11, 27, 8, 17, 7, 6, 9, 4, 8, 7, and 13 respectively. One of the primers for each locus was labeled with fluorescent dye to generate fluorescent PCR products, which were separated on an AB™ 3130 DNA Analyzer to allow genetic data to be collected. From these data: the mean of heterozygosity was found to be 0.7145; the polymorphism information content ranged from 0.461 to 0.883; and, with the exception of locus DXS6807, the X-STR loci were found to be highly polymorphic as their polymorphism information content was higher than 0.6. Males were studied to determine the

polymorphic nature of a single allele for these X-STR loci, resulting in the observation of 129 haplotypes from 130 unrelated males. The Logarithm Of Odds (LOD) score test of pairwise linkage study using the LINKAGE program ranged from 4.4 to 23.73; this indicated that these 11 loci are highly linked, as would be expected. Based on a linkage study using 47 families, all with at least with two children, all 11 loci were inherited as if linked, except in one case where a crossover was observed in the subgroup of DXS680702/DXS680703/DXS680704. The maximum likelihood estimation of the recombination rates were found to be all less than 0.01. Since X-STR loci have been proven to be powerful in solving particular kinship cases, the haplotype of linked X-STR loci could provide more discrimination power. These newly established 11 clustered X-STR loci are highly polymorphic and have the potential to aid in kinship testing where X chromosome loci currently play a role.

X-STR Loci, Linkage, Kinship Test

A44 Evaluation of Sampling Methods for the Successful STR Typing of Dreadlocked Hair Samples

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The goal of this presentation is to offer attendees an evaluation of methods used to obtain DNA from atypical hair samples, namely dreadlocks.

This presentation will impact the forensic science community by offering methods of DNA sampling procedures when nontraditional hair evidence, such as dreadlocks, is analyzed.

Hair evidence is often found at crime scenes and submitted to forensic laboratories for DNA analysis. Typically, the type of hair evidence submitted has been individual hair strands. However, with the increased popularity of natural hairstyles, dreadlock hair may be submitted for casework analysis as well. In a review of the literature, no instances of DNA analysis of dreadlocked/locked hair samples were found. The sampling methods to obtain an Short Tandem Repeat (STR) DNA profile from locked hair samples were evaluated. The case study presented here is one in which a dreadlock was recovered at the scene of a homicide and also the resulting bench study.

Dreadlocks, also called locks, are formed by allowing the hair to shed into itself, tangle, and mat. The thickness, as well as the length, of the matted hair can vary among individuals. STR DNA typing of hair evidence is traditionally accomplished by isolating the biological evidence from the root of individual hair strands. Due to the composite nature of dreadlocks, it may be possible to perform nuclear DNA testing even when a visible root tag is lacking. Two methods of sampling dreadlock/lock hair for STR typing will be discussed in this presentation. The first sampling method utilizes a cut portion of the actual dreadlock. This method is independent of the width of the dreadlock. The second method utilizes the wet swab technique. Use of the swab technique does depend on the width of the lock.

Sampling methods were performed on 18 dreadlock/lock samples. Each sample was externally swabbed and, depending on thickness, cross-sectioned and internally swabbed with a single wet cotton tip swab. Additionally, cuttings of the proximal, middle, and distal portions of the lock were collected for DNA typing. A modified EZ1® extraction protocol for the pretreatment of hair was utilized for the cuttings. STR typing was performed using Life Technologies® AmpFLSTR® Identifiler® PCR Amplification Kit.

The researchers will present a case study as an example of the practical application of the two sampling methods.

STR Typing, Hair Evidence, DNA

A45 Improving Physical Evidence Storage and Efficiency of Downstream Processing of DNA in Forensic DNA Analysis

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After attending this presentation, attendees will gain an understanding of how osmolytes can be utilized to improve the storage and downstream analysis of DNA.

This presentation will impact the forensic science community by providing data on the use of organic osmolytes in improving the storage and analysis of DNA in forensics. This presentation will enhance research in the areas of preservation and analysis of forensic DNA analysis.

DNA collection and analysis have provided the forensic science field with a powerful tool for convicting the guilty and exonerating the innocent. For DNA analysis, the forensic field utilizes the Short Tandem Repeated (STR) DNA sequences which are widespread throughout the human genome. The forensic DNA community has adopted tetranucleotide repeats, which may be amplified using the Polymerase Chain Reaction (PCR) with greater fidelity and fewer stutter artifacts than other repeats. DNA analysis does have its drawbacks including inhibition, degradation, and artifacts present in the analysis. The use of osmolytes in circumventing some of these obstacles is explored in this study. In a previous study, DNA from blood stains incubated with trehalose were found to be less degraded when preserved at room temperature or 90°C.¹

Organic osmolytes are small solutes that stabilize macromolecules and cells utilize them to counteract denaturation.^{2,3} Similar compounds are accumulated by some organisms in anhydrobiotic, thermal, and pressure stress environments. Among the major groups of osmolytes identified are polyols, free amino acids and their derivatives, and methylamines and methylsulfonium compounds. Osmolytes have been found to engage in unique reactions that can protect cells in different ways. Osmolytes are naturally produced by organisms that have adapted to extreme conditions such as high temperature, low humidity, and high salinity. These compounds have previously been shown to increase the thermal stability of proteins under stressful conditions.

It is possible that forensic evidence must be maintained for many years as the backlog in casework samples is eliminated. Additionally, evidence must be retained for potential re-examination (for example, by the defense lab as well as for possible future analysis using new technologies). The cost to retain evidence in freezer space can be significant. If effective preservatives could be added to the biological evidence, costs could be reduced by storing the evidence at room temperature. To encourage suitable preservation and storage of forensic evidence, the National Institute of Justice (NIJ) recommends determining cost-effective methods for storage of evidence.

The present project studied the utility of additives (organic osmolytes) on the long-term storage of DNA from biological samples as well as in improving the downstream processing of DNA in forensic samples by preventing formation of stutter artifacts during PCR. To assess the ability of preservatives to improve the storage of biological samples, osmolytes (myoinositol, trehalose, raffinose,

and stachyose) were incubated with DNA samples extracted from biological fluids for various time periods and extreme environmental conditions (e.g., high temperature, humidity). DNA from these samples was then analyzed by STR analysis. Moreover, the results show that polyols containing four and five carbons (erythritol and xylitol) did not provide the same protection as polyols with at least six carbons capable of forming a ring (e.g., myoinositol, trehalose, and raffinose). It was also determined that these osmolytes can protect DNA from DNase I treatment used to simulate DNA degradation. Additionally, the utility of osmolytes to reduce the stutter artifact in DNA analysis was investigated. Osmolytes (sorbitol and betaine) were found to decrease stutter products in microsatellite analysis by 50% compared with normal unmodified PCR conditions. These studies provide useful modifications to forensic DNA analysis of STRs.

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Osmolytes, DNA Preservation, DNA Analysis

A46 Changes in DNA After Bone Marrow Transplant: Challenge for Forensic Genetics

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After attending this presentation, attendees will be informed about novel forensic situations in which blood, buccal swabs, and hair follicles of humans are not a reliable source for individualization using DNA profiling techniques.

This presentation will impact the forensic science community by highlighting the very important concept of chimerism in humans. If not considered properly, this can lead to false results when DNA profiling techniques are used for identification purposes.

The role of forensic genetics within the investigative process is to compare a sample recovered from a crime scene with a suspect. Many individuals are receiving hematopoietic stem cell transplantation (bone marrow/peripheral blood, or umbilical cord blood transplant) from HLA matched or unrelated donor. Bone Marrow Transplant (BMT)/Peripheral Blood Stem Cell Transplant (PBSCT) has been widely accepted as a convincing life-saving treatment modality for the various malignant and non-malignant hematologic diseases. This has the potential to change the genetic makeup and blood group of an individual, posing a challenge to forensic scientist's to solve the case successfully. This challenge results because if a subject has a history of successful allogenic bone marrow transplantation, donor cells would be present in recipient blood cells and consequently, blood would not be a suitable substrate for personal identification or kinship study. This situation is called chimerism and refers to the presence of lymphohematopoietic cells of non-host origin. These cells could be derived incidentally from a fetal-maternal transfusion, a blood transfusion, or purposefully after hematopoietic stem

cell transplantation. Such genetic peculiarity may prevent the association of the perpetrator of an offense with the stain left at the crime scene or lead to false paternity exclusions. Forensic scientists may encounter the problem of mixed or completely mismatched DNA profiles of a single individual when the source of the biological material being analyzed is from a person who is a genetic chimera. The goals of this study were to evaluate the chimerism level in different biological specimens and to identify the ideal specimen for individualization using DNA profiling techniques. Three different biological specimens (peripheral blood, buccal swab, and hair) were analyzed at five different time intervals (pre-transplant, +21 days, +3 months, +6 months, and +1 year) from 25 different recipients of Hematopoietic Stem Cell Transplantation (HSCT) to estimate the chimeric status of patients. Written consent was taken from every participant prior to sampling. Genomic DNA was extracted by organic extraction method. Out of 25, 13 patients had severe aplastic anemia, six patients had thalassemia, five patients had myelogenous leukemia (acute myeloid leukemia or chronic myeloid leukemia), and one patient had RBC aplasia. Eighteen patients received PBSCT while seven patients underwent Allo- BMT. Out of 25, 12 patients were sex mismatched with their respective donors while 13 were not. Used were 16 different STR markers (AmpFtSTR identifier kit) to generate the genetic profile of the individuals. Peak areas of donor and recipient alleles were used to calculate the chimerism percentage. Donor chimerism was observed in blood (93-100%), buccal swabs (2-57%), and hair follicles (0%). In the blood samples, 19 out of 25 patients showed complete donor chimerism (100%). Three patients showed chimerism with a range of 92.4-100% and three patients showed mixed chimerism with a range of 46.9-97% at various time points. Buccal swabs showed donor chimerism in all the samples at all the time points. Eight out of 25 patients showed donor chimerism below 10%. No donor chimerism was observed in hair follicle samples. Standard deviation and coefficient of variation was calculated for every sample at every time point and the results suggested that blood and buccal swabs are not suitable for personal identification in chimeric patients. Hair, however, can be used as a good source of biological material to obtain the original genotype profile of an individual.

Bone Marrow Transplant, DNA Profiling Technique, Chimerism in Humans

A47 Examination of Nonoxynol-9, Including an Investigation of Interfering Compounds and Potential False Positives

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After attending this presentation, attendees will understand and be able to implement a method for the analysis of Nonoxynol-9 (N9), a common spermicide. Additionally, attendees will have a broad understanding of the specificity of this method and the potential sources of false positives.

This presentation will impact the forensic science community by providing a highly specific methodology for the analysis of a common spermicide as well as a context for the interpretation of results as applied to forensic analyses in sexual assault cases.

Examination of items of evidence from sexual assault crimes may, in some instances, require the detection of nonoxynol-9, a spermicidal compound. Nonoxynol-9 is a surfactant compound which is often found in condoms and sexual lubricants. The identification of this material can act as valuable forensic evidence,

providing a trace compound which can be used to link a suspect and victim to used condoms or sexual lubricants which may have been employed in the incident.

A combination of techniques can be used to obtain structural and mass information regarding nonoxynol-9. The most common techniques for the analysis of nonoxynol-9 include Fourier Transform Infrared Spectroscopy (FTIR) and Direct Analysis in Real Time-Mass Spectrometry (DART®/MS). Structural information of this polymeric compound can be elucidated from FTIR or through characteristic fragmentation ions when analyzed by DART®/MS. Also, by providing a softer ionization in the MS analysis, the protonated molecular ion can be detected, identifying the presence of the nonoxynol-9 molecule specifically.

In addition to the development of a methodology for the analysis of nonoxynol-9, several potential interfering materials were analyzed to determine potential sources of false positives. Common cosmetic products which may be present as background materials in forensic science casework (such as lotions) were analyzed. Additionally, as nonoxynol-9 is a surfactant, other surfactants were analyzed to determine whether other surfactant materials could provide false positives. Surfactants examined included dish soaps, laundry detergents, stain removers, and body soaps. Finally, other sexual lubricant materials were examined to determine if the ingredients in commercial condom and sexual lubricants would interfere with the analysis of nonoxynol-9. A robust sample set of interfering materials was tested during this study in order to evaluate the specificity of the analysis, covering over 100 various commercial products.

Some materials were found to have molecular masses which were consistent with peaks associated with the nonoxynol polymer, while other materials were found to be structurally similar; however, very few commercial products produced positive results for both the fragmentation pattern and the molecular ion of nonoxynol-9. This supports the high degree of specificity of the analytical scheme. Anomalous and outlier results will be discussed in detail.

The implementation of this methodology will prove valuable to forensic science service providers involved in performing analyses for cases requiring the detection of spermicides in sexual assault cases. Not only will a discussion of the analytical scheme be provided, but the context with which analytical results can be interpreted will be discussed.

The opinions or assertions contained herein are the private views of the author and are not to be construed as official or as reflecting the views of the Department of the Army or the Department of Defense.

Spermicide, Sexual Assaults, Trace Evidence

A48 4N6FLOQSWABS™: An Alternative for Sample Collection

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After attending this presentation, attendees will understand the design of the Copan 4N6FLOQSWABS™, the swab's advantages and disadvantages when compared to cotton swabs, their use in conjunction with the Nucleic Acid Optimizer, and the DNA yield from the extraction of samples collected with Copan 4N6FLOQSWABS™. Attendees will also understand the design and functionality of a Nucleic Acid Optimizer (NAO™ Baskets) in DNA processing and the advantages and disadvantages of the use of NAOs.

This presentation will impact the forensic science community by presenting the 4N6FLOQSWABS™ as a sample collection method that can be used as an alternative to the cotton swab, which is used commonly to collect DNA evidence at crime scenes and in forensic laboratories.

4N6FLOQSwabs™ are made of parallel short nylon strands that are flocked onto a plastic stick lacking an inner core that can trap cellular materials. This is in contrast to typical cotton swabs with a mattress design that are highly absorbent and known to trap cellular materials in their core. The swab is also designed to neutralize microbial contaminants, while preserving the integrity of nucleic acids, without the need to dry the swab. The design of the Nucleic Acid Optimizer is different from the typical spin basket with a woven bottom. Instead of a woven plastic construction, the bottom of the NAO™ Basket is plastic with "valves" that do not allow liquid to pass through under normal conditions. The semi-permeable spin basket allows the elution of liquid through the valves upon centrifugation at maximum speed. This design allows for the incubation of a sample substrate to occur directly inside of the spin basket, without the loss of buffer through the valves. Upon direct centrifugation, the filtrate can be collected, which makes the transfer of substrate and buffer unnecessary after incubation. By reducing the number of transfer steps, the chances of sample contamination could be reduced, while DNA yield could possibly be improved.

Due to its design, it has been proposed that 4N6FLOQSwabs™ are more effective at releasing cellular materials than cotton swabs, thus the recovery of DNA from samples can be improved with the use of the Copan 4N6FLOQSwabs™ used in conjunction with the NAO™ Baskets when compared to cotton swabs extracted with NAOs. To test this, DNA was extracted from blood samples that were spotted on glass and swabbed with 4N6FLOQSwabs™, with conventional forensic cotton swabs, and from blood samples spiked directly onto both swabs. For each condition, six replicate blood samples were prepared and extracted. Samples were also extracted two weeks later using the NAO™ Baskets during the lysis incubation stage.

When blood was spotted on glass, there was, on average, a 52% increase in DNA recovery when swabbed with 4N6FLOQSwabs™ compared to swabbing with cotton swabs. The average recovery for the 4N6FLOQSwabs™ was $1.206 \pm 0.21\text{ng}$ compared to $0.795 \pm 0.31\text{ng}$ recovered by the cotton swabs on glass. Furthermore, when blood was spiked directly on the swab, there was, on average, a 196% increase in DNA recovery when spiked on 4N6FLOQSwabs™ as compared to cotton swabs. The average recovery directly on the swab for the 4N6FLOQSwabs™ was $0.880 \pm 0.09\text{ng}$ compared to $0.297 \pm 0.11\text{ng}$ recovered from the cotton swabs.

Results showed that the 4N6FLOQSwabs™ provided the best DNA recovery overall. Furthermore, the increase in the DNA recovery with 4N6FLOQSWABS™ when blood was spiked directly onto the swabs shows that the 4N6FLOQSWABS™ are more effective at releasing cellular materials from within the swab.

4N6FLOQSwabs™, Nucleic Acid Optimizer Baskets, Improved DNA Sample Release

A49 DNA Typing Compatibility With a Rapid, One-Step Saliva Screening Test: Phadebas® Forensic Press Test

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The goal of this presentation is to inform attendees of the benefits and limitations of the Phadebas® Forensic Press (PFP) test. Furthermore, attendees will understand the best methods for extracting DNA from substrates tested with Phadebas® paper.

This presentation will impact the forensic science community by providing valuable information about a saliva screening technology that can be employed in forensic laboratories for more effective, rapid presumptive testing of biological fluids.

DNA typing has become an important tool in forensic science. Steps in forensic DNA typing include screening for biological samples such as blood, saliva, or semen; DNA extraction; quantification using quantitative Polymerase Chain Reaction (PCR); PCR amplification of genetic markers such as autosomal or Y chromosome short tandem repeats; comparison to reference or database DNA profiles; and then interpretation and reporting of results.

Screening a substrate for bodily fluids is an extremely important step for locating areas that may contain DNA. Several different methods have been developed for saliva.¹ The PFP test is a presumptive saliva test that utilizes a preloaded paper that will react with the enzyme amylase, a component of saliva.^{2,5} Because of its ability to screen for amylase while simultaneously locating stains, the PFP may prove to be an effective, rapid method for screening; however, it is important to assess whether the PFP introduces any inhibitors to downstream processing such as PCR amplification.⁶

Based on previous studies, it is hypothesized that the PFP will provide a rapid and sensitive method for locating multiple saliva stains simultaneously without introducing inhibitors to DNA profiling. To evaluate sensitivity of the method, dilutions of saliva (from neat down to 1:5,000) were stained onto cotton substrates. Saliva dilutions from six different donors were prepared as described and then tested using the PFP and visual detection for up to 40 minutes. In addition, replicate saliva samples were prepared and then subjected to UV degradation to evaluate sensitivity on environmentally abused stains. Mixtures of blood:saliva and semen:saliva were prepared (1:5, 1:2, 1:1, 2:1 and 5:1) and stained on cotton substrates. All of the samples were then processed with the PFP as previously described.^{2,3} The fabric containing the stains and the PFP paper were then subjected to organic DNA extraction, DNA quantification (Quantifiler® Duo qPCR kit), and PCR amplification (Identifiler® Plus and Yfiler® STR kits) to evaluate whether any inhibition was observed.

The PFP limit of detection was observed between 1:10 to 1:1,000 for saliva dilutions. Degraded samples exhibited delayed positive results taking approximately twice as long to detect (e.g., neat control saliva detected in 4min 42sec and UV-treated neat saliva in 7min 57sec). Control saliva was detected in just under five minutes whereas UV cross-linked neat saliva was detected in just under eight minutes.

For mixtures, all ratios of blood and saliva were detected but reactions were delayed out to 30 minutes for the lowest ratio of saliva to blood (1:5). All ratios of semen and saliva were detected in less than 10 minutes. Neat semen stains were not detected with the PFP; however, AP testing showed positive results.

Full STR profiles for both Identifiler® Plus and Y Filer® were amplified using DNA extracted from cuttings of the fabric or cuttings of the fabric and PFP paper for all saliva dilutions down to 1:100. Additionally, full profiles were also developed using DNA extracted from the fabric substrate for all mixtures of semen and saliva with no apparent inhibition. DNA extracted from the PFP paper provided full STR results only for the neat and 1:2 dilutions. No additional alleles were amplified when combining DNA extracted

from the PFP paper.

Amplification of full STR profiles for both Identifiler® Plus and Yfiler® multiplexes were observed using DNA extracted from cuttings of the fabric or cuttings of the fabric and PFP paper for all saliva dilutions down to 1:100. Additionally, full profiles were also observed using DNA extracted from the fabric substrate for all mixtures of semen and saliva with no apparent inhibition. DNA extracted from the PFP paper provided full STR results only for the neat and 1:2 dilutions. No additional alleles were amplified when combining DNA extracted from the PFP paper.

Preliminary results support the hypothesis — the PFP appears to be a rapid, sensitive, method capable of detecting multiple stains simultaneously without inhibiting PCR. Additional tests and validation will be performed to determine the efficacy and limitations of the Phadebas® Forensic Press test.

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PCR, Phadebas® Forensic Press Test, Saliva Screening

A50 Preservation of Human Tissue Samples in Tropical Climates

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After attending this presentation, attendees will be presented with the results of research comparing the efficiency of four solutions to preserve DNA in human cadaveric skin and muscle samples when stored in simulated tropical conditions for up to six weeks.

This presentation will impact the forensic science community by providing an alternate sampling approach when faced with an overwhelming number of victims to identify in the aftermath of a mass disaster.

One of the most important and immediate considerations after a mass fatality event is victim identification. In these circumstances, forensic personnel may be faced with the task of identifying hundreds or even thousands of human remains. The Disaster Victim Identification (DVI) response by local and international agencies is often complicated and delayed by

remote locations, harsh environmental conditions, and lack of resources. In addition, electricity and communication networks are often compromised and adequate freezer facilities to house the victims may not exist. Therefore, large numbers of bodies decompose rapidly in hot and humid conditions which leads to serious biohazard problems; however, from a DVI perspective, the DNA in those tissues is also degrading which makes genotyping more difficult with time. In these circumstances, the use of a simple field preservative to quickly halt DNA degradation and store large numbers of tissue samples at ambient temperature prior to genotyping would be a valuable tool.

This pilot project evaluated how well three in-house preservatives (combinations of TENT buffer, ETOH, and salts) and one commercial solution (DNAGard® Tissue, Biomatrix®) protected the DNA in fresh cadaveric skin and muscle samples (n= 28 each) at 35°C in humid (40-50% humidity) conditions for up to six weeks. In addition, the quantity and quality of DNA leeching from these tissues into the surrounding solution was also assessed. The quantity of DNA in both the tissue samples and the “free” DNA in the preservative was determined using quantitative real-time PCR and the quality of the DNA was assessed using the Powerplex® 16 HS STR amplification kit. The “free” DNA in solution is available for direct purification and therefore avoids the much longer process of extracting DNA from the tissue itself.

The results of this study demonstrate that DNA was preserved in both muscle and skin samples using all four preservatives tested. The TENT buffer generated the lowest DNA yields (5ng/μL) and the ethanol-based solution yielded the most DNA (120ng/μL) from both tissues. This presentation will also present data showing that adequate amounts of DNA (2-40ng/μL) leached into solution for direct DNA isolation and genotyping with all preservatives except the ethanol-based solution. However, complete STR profiles were only obtained from the DNAGard® and modified (increased NaCl) TENT solutions, suggesting that these two tissue preservatives are better at protecting the “free” DNA in solution from further damage and/or degradation than the other preservatives tested.

By maximizing the quantity and quality of “free” DNA in the preservative solution, not only could samples be stored adequately at ambient temperatures, but the time-consuming steps of tissue digestion and DNA extraction may be eliminated, allowing for higher throughput of samples for identification.

DNA Preservation, Disaster Victim Identification, DNAGard® Tissue

A51 Characterizing Rates of Allelic Dropout and the Impact on Estimating the Number of Contributors

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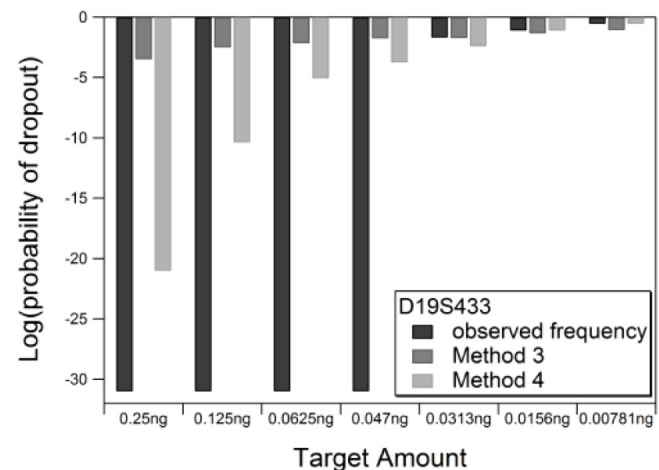
After attending this presentation, attendees will understand the impact different methods of modeling allelic dropout in forensic DNA samples has on estimating the number of contributors (n_c) to a DNA sample.

This presentation will impact the forensic science community by demonstrating ways to characterize allelic dropout — one essential piece of information that influences estimating the number of contributors to a forensic DNA sample.

Allelic dropout affects the weight an item of evidence will carry in court. In particular, it may affect the assessment of how many individuals contributed to the evidence. Accurately determining n_c is of import since an assumption regarding n_c is made when calculating the likelihood ratio, which compares the hypothesis of the prosecution and the hypothesis of the defense.

Five methods to characterize the probability of allelic dropout ($Pr(D)$) were compared against real data collected from 96 known, single-source samples. These samples were amplified at seven target amounts, injected for 10sec and analyzed using the Local Southern method which uses an analytical threshold of 1 RFU. The impact two of the five methods had on estimating n_c to a DNA sample was also evaluated.

The first four methods investigated the ability to predict allelic dropout based on single-source calibration data which determined the distribution of peak heights of alleles at a specific target mass and locus. Probability Density Functions (PDFs) of peak height were determined by fitting the observed heights to a weighted Gaussian (Method 1), and a non-weighted Gaussian curve (Method 2). Method 3 was slightly different in that the data was not fitted. In this instance, the sample mean and standard deviation of the observed heights were calculated and utilized to generate the PDF. Given PDFs of the peak height, $Pr(D)$ was estimated as the probability of observing a peak height <1. Since a logistic regression to characterize allelic dropout has previously been investigated in other studies, a logistic regression (Method 4) was used to fit the observed frequencies of dropout of the calibration data versus the target amount per locus.¹ An exponential regression (Method 5) for characterizing dropout was also explored.



The resultant probabilities were then compared to the observed frequencies of dropout to determine which method accurately predicts the rates of allelic dropout. While Method 3 resulted in good approximations of dropout rates at most targets, of all five methods tested, Method 4 resulted in the most accurate dropout approximations over most loci at most target masses tested. The estimated $Pr(D)$ for Methods 3 and 4 and the observed dropout frequencies for a representative locus is displayed.

z	APP(i)=Pr(n _c =i E)					
	n _c =1			n _c =2		
	Observed	Method 3	Method 4	Observed	Method 3	Method 4
0.25	1.00E+00	1.00E+00	1.00E+00	1.31E-10	1.90E-10	1.31E-10
0.125	1.00E+00	1.00E+00	1.00E+00	1.56E-10	2.49E-10	1.31E-10
0.063	1.00E+00	1.00E+00	1.00E+00	1.73E-10	1.05E-09	1.35E-10
0.047	1.00E+00	1.00E+00	1.00E+00	2.42E-10	1.13E-09	1.56E-10
0.031	1.00E+00	1.00E+00	1.00E+00	7.65E-10	4.33E-09	3.96E-10
0.016	1.00E+00	1.00E+00	1.00E+00	5.05E-09	2.68E-09	6.34E-09
0.008	1.00E+00	1.00E+00	1.00E+00	5.09E-05	2.27E-09	4.82E-05

Further, the impact of using Methods 3 and 4 on estimating n_c to a DNA sample containing no more than two alleles per locus was determined by calculating the *A Posteriori Probability* (APP) for $n_c=1,2,3,4$ where $APP(i)=Pr(n_c=i|E)$. Here the APP is determined by considering all genotype combinations (with and without dropout) that could have resulted in the observed signal. An $APP(i)=1$ signifies that, given the evidence, i is the most likely value for n_c . The APPs using $Pr(D)$ according to Methods 3 and 4 were then compared to the APPs using the observed dropout frequencies. For the 50 single-source samples tested, both Methods 3 and 4 resulted in 100% of the single source profiles identified as such. The accurate identification of $n_c=1$ occurred even at extreme low-template masses, where the observed frequency of dropout was as high as 0.39. Also, the APPs are closest to 1 when $n_c=1$ and as n_c increases, the APPs decrease, as expected. Representative values of the APPs for $n_c=1,2$ are displayed in the table. Further studies will evaluate the impact these dropout models have on samples that contain greater than two alleles at a locus.

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Allelic Dropout, Number of Contributors, Forensic DNA Analysis

A52 An Improved Method of DNA Extraction From Environmentally Challenged Teeth

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After attending this presentation, attendees will be knowledgeable concerning research investigating an improved method for retrieving DNA from teeth exposed to various environmental conditions. The research explores an alternative method of dentine removal from teeth that improves DNA yield and STR success while retaining tooth morphology.

This presentation will impact the forensic science community by providing improved methods for retrieving high-

quality DNA from environmentally challenged teeth for victim identification.

Often in cases such as mass disasters, war remains, missing persons, acts of terror, and airplane crashes, only skeletal remains are recovered for victim identification. The ability to extract high-quantity and quality DNA from bone and teeth is important for identification; however, common methods for DNA extraction from teeth involve various levels of tooth destruction, which limits, or even eliminates, the possibility of further morphological analysis. In addition, only non-invasive methods are permitted on some samples, such as ancient remains housed in museums. By using methods that do not solely target the DNA-rich areas of teeth, PCR inhibitors are often co-extracted with the DNA and interfere with genotyping.

Molars (n=14) sourced from seven individuals (two molars each) were used during this study. Each set of teeth were exposed to one of seven treatments: buried for 12 or 24 months, surface burial for 12 or 24 months, saltwater for six months, freshwater for two months, or fire. One tooth from each pair was prepared for DNA extraction by removing the entire root system and grinding it into a powder using a freezer mill, while the other tooth was prepared using a filing method. This technique involved using endodontic files to scrape the inside of the root canals and pulp chamber without piercing the enamel crown or destroying the root structure.

The quantity and quality of DNA yielded from the teeth using these two techniques was compared. DNA was quantified using a real-time quantitative PCR assay and DNA quality was assessed using the Powerplex® ESI 16 STR amplification kit. Although less dentine powder was generated using the filing method, more amplifiable DNA was recovered in the majority of cases. Moreover, when the DNA yield-per-milligram of powder (efficiency) was calculated, the filing method generated 1.5- to 35.5-fold more amplifiable DNA. The filing method also generated more complete, balanced, and consistent STR profiles than the whole root method across the sample set. These data suggest that the filing method is superior to processing the entire root for STR profiling DNA from teeth exposed to various environmental insults.

Teeth, DNA Extraction, Victim Identification

A53 Inter-Laboratory Testing of a Highly Sensitive Quantification System for Assessing DNA Quality in Forensic Samples

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After attending this presentation, attendees will have a better understanding of a new Innoquant H Kit from InnoGenomics Technologies and how the new qPCR, which utilizes two independent genomic targets, is able to provide information on sample degradation and sample inhibition. Attendees will learn about the capabilities of the kit on inhibited and degraded samples using a modified thermal cycling extension protocol and different-

sized internal positive controls.

This presentation will impact the forensic science community by demonstrating the effectiveness of the Innoquant H Kit and its abilities to work with degraded DNA as well as DNA samples containing inhibitors. With efficiency for both the long and short targets being consistently above 90%, forensic laboratories will be able to use this kit in order to analyze highly compromised samples.

Real-time PCR quantification of human DNA can provide an important estimate of the amplifiable DNA in a biological sample. Currently, methods being utilized in forensic DNA laboratories include SYBR[®] Green, Plexor[®] HY, and Quantifiler[®] Duo TaqMan assays.¹⁻³ The recent advances in mini-STR analysis systems have now made it possible to analyze highly compromised samples.

A quantification system that estimates the level of degradation in a forensic sample will be a useful tool for DNA analysts. There are previous reports of systems that provide a quality assessment of degraded DNA samples. One uses a Ya5-lineage *Alu* genetic element and a second uses a multiplex simultaneously assessing nuclear and Y chromosome targets ranging from 67bp to 190bp.^{6,5} The advantage of an *Alu* system is the presence of a large number of fixed insertions. It has been reported that only 20% of the Yb-lineage *Alu* elements are polymorphic for insertion presence or absence in the human genome.⁴ A large number of these fixed elements are present in every human genome, enhancing sensitivity and minimizing the individual specific variation possible when using a multi-copy target quantification system.

A multi-copy intra *Alu* based approach to quantify human specific DNA in an evidence sample has been successfully used to obtain DNA quantification with high sensitivity.⁷ *Alus* are Short Interspersed Elements (SINE), approximately 300bp insertions, which are distributed throughout the human genome in large copy number. The use of an internal primer to amplify a segment of an *Alu* element allows for higher primate specificity as well as high sensitivity when compared to a single copy target.

The new qPCR utilizes two independent genomic targets. Primers and TaqMan probes were designed using two independent intra retrotransposon insertions targets. The 80bp "short" target sequence is from an *Alu* insertion whereas the 290bp "long" target sequence is from a separate retrotransposon element. The primers and probes for the two targets are selected such that they have no interaction among themselves and are completely independent. The ratio of the quantity of long targets versus short targets provides a useful assessment of the quality of DNA. This quality ratio can have applications in predicting the profiling success of forensic samples. The use of a synthetic target as an Internal Positive Control (IPC) provides an additional assessment for the presence of PCR inhibitors in the test sample.

Inter-laboratory testing of different-sized IPCs and results on modifying thermal cycling parameters including additional extension times during qPCR on standard DNA, degraded DNA, and inhibitor-spiked DNA will be reported. Additional results on the developmental validation studies for this system will be presented including multiplexing PCR, degradation, and inhibition. The quality ratio will also be evaluated for the ability to predict profiling success on physically degraded DNA samples.

Preliminary results demonstrate that the efficiency for both the long and short targets is consistently above 90%. The amount of synthetic IPC target was adjusted to provide reproducible Ct values between 18-22 cycles for samples with no inhibition. Precision and sensitivity studies indicated that this system has a sensitivity threshold in the range of 3-4pg, similar to those reported for other *Alu*-based quantification systems. Studies comparing this system with other commercially available quantitation systems show concordance of quantitation values between systems. Furthermore, the preliminary inter-laboratory results demonstrate

the predictive value of the quality ratio on degraded DNA, and the IPC results on inhibitor spiked samples.

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qPCR, Degraded DNA, PCR

A54 Analysis of Stochastic Level DNA Mixtures

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After attending this presentation, attendees will have a better understanding of the behavior of major and minor contributors as they approach and then drop below the stochastic level in a DNA mixture. This study presents data regarding the peak height ratio relationship between two-person mixture profiles in various quantities and mixture ratios above and below the stochastic level. It also presents data comparing the results of mixed stochastic level samples processed using the Promega PowerPlex[®] 16 HS Kit and the Promega PowerPlex[®] Fusion Kit.

This presentation will impact the forensic science community by providing further experimental data regarding how allele peak heights relate to each other, both between contributors to a mixture and among heterozygous sister pairs. In particular, these relationships become difficult to predict as the mass of each contributor approaches the stochastic threshold at different times due to the disparate starting quantities of the major and minor contributors. Ultimately, this data will assist analysts with the interpretation of mixed samples in which one or both of the

contributors are present in a stochastic quantity.

Analysis of mixed DNA samples poses a significant challenge to the forensic science community. The most common mixture samples seen are those with two contributors. Difficulties with interpretation start to arise when the quantity of these samples are low. Interpretation is also challenged by the difference in the quantities of the respective contributors (i.e., the ratio that they are mixed in). The stochastic effects include allele drop-out, allele drop-in, and heterozygous peak imbalance. A heterozygous allele peak height ratio was initially calculated. The peak height ratio was then used as a guide to study the relationship of the heterozygous peak height imbalance between and among the major and minor contributors. With these combined factors, this study examined at what point in the ratio and the amount of the samples these stochastic effects were likely to be seen.

The study showed the peak height relationships between the major and minor contributors in various ratios and dilution factors to include a broad range to observe the stochastic effects at multiple levels. Mixture samples were created in ratio series of 1:2, 1:3, and 1:5 and each of the contributors were mixed as a major or a minor contributor in a series of experimental samples. The prepared samples were amplified using the Promega PowerPlex® 16 HS Kit. Data collected demonstrated the peak height level at which the minor component heterozygous sister alleles began to show imbalance. Further data indicated the point at which minor component alleles dropped out and major component heterozygous pairs began to show imbalance. This data was compared to the same samples processed individually in order to examine if the stochastic threshold differed between the mixed samples and the single source samples. The results indicated that minor contributors began to show heterozygous peak imbalance consistent with stochastic level profiles at higher concentrations of input DNA and at higher peak heights than in the single source samples. For this reason, it may be advisable to increase the stochastic threshold used to interpret mixed STR results over that used for single-source samples. The same samples were then processed using the Promega PowerPlex® Fusion kit for a comparative analysis with the Promega PowerPlex® 16 HS kit. The extra loci included in the new kit were analyzed to see if they were beneficial for the interpretation of stochastic-level DNA mixture samples. The increased number of loci made the separation of mixed profiles easier overall due to the larger number of available alleles. The smallest loci (D3S1358, D16S539, TH01, and D8S1179) proved to be less prone to stochastic level peak height imbalance, also easing interpretation by providing clearer separation between contributors.

Stochastic, DNA, Mixtures

A55 DNA Studies in Cases of Child Sexual Abuse

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The goal of this presentation is to characterize cases of child sexual abuse in which biological samples were collected for DNA studies in order to understand: how long it is possible to find DNA evidence after the abuse, the proportion of samples that yield positive results, the influence of the victim's age, the presence of physical injury, and the type of practices in DNA findings.

This presentation will impact the forensic science community by clarifying how collection of biological samples for DNA identification should be performed in cases in which the time

lapse between the last contact and the exam is less than 72 hours, despite, in specific cases, collection being done beyond that time frame and in the presence of normal or nonspecific findings.

Child sexual abuse is frequently associated with normal or nonspecific physical findings. Biological evidence may be the only evidence of abuse; therefore, having serious implications on the perpetrator's conviction or child protection measures.

Guidelines from the American Academy of Pediatrics recommend considering forensic evidence collection for up to 72 hours after the alleged assault. Studies regarding the time of spermatozoa survival after sexual intercourse in adult women have supported this timeframe; however, several experts reported few or no positive DNA results when the forensic medical examination takes place more than 24-hours after the sexual contact. Nevertheless, none of these studies included DNA amplification techniques which have higher sensitivity. Therefore, the forensic medical practitioner should be aware of the rapid advance of DNA identification techniques and consider evidence collection on a case-by-case basis.

A retrospective study was conducted based on the analysis of 231 reported cases of suspected sexual abuse of victims younger than 18 years old that were observed at the Portuguese National Institute of Legal Medicine (INML) – North Branch (Porto) from 2004 to 2011, in which samples were collected for DNA identification.

The selected cases correspond to 25.4% (n=231) of the 911 suspected cases of child sexual abuse. In 79.7% of these cases, the victims were female and 85.3% of the abusers were male. Identifiable DNA was found in 59 cases (25.5%); nine in children younger than ten years old; and, in 35 cases, less than 24 hours after the alleged abuse. In three cases, DNA was found in the victims' clothes, despite sample collection that had occurred over 72 hours after the abuse. The proportion of positive DNA results was higher for samples collected from the clothes (30%) than for those collected from the victim's body (14%). A significant correlation was found between the presence of identifiable DNA and physical injury (p=0.016), a shorter interval of time between the abuse and sample collection (p=0.000), and older victim's age (p=0.001, t=3.388).

Forensic medical examination should be performed whenever there is a suspicion of child sexual abuse and include collection of biological samples for DNA identification in all cases in which the time between the last contact and the exam justifies it, as well as in the presence of normal or nonspecific findings. In specific cases, sample collection may be performed beyond the currently recommended limit of 72-hours.

The increasing sensitivity of DNA tests is likely to continue to extend current time limits for evidence collection.

Further prospective studies are needed, controlling for variables in which information is usually absent.

Child Sexual Abuse, DNA, Forensic Medical Examination

A56 Development and Evaluation of an Objective Method for Human Hair Examination Using Image and Mitochondrial DNA Analyses

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After attending this presentation, attendees will understand the potential of a fusion-based approach based on the conjunction of image analysis and mitochondrial DNA analysis as an objective methodology for discriminating human hair samples. This approach will feature the conjunction of image analysis and mitochondrial DNA analysis.

This presentation will impact the forensic science community by providing an objective method for the forensic examination of human hair samples that cannot be successfully subjected to nuclear DNA analysis. The complementarities of the two proposed methods yields a higher power of discrimination than either method alone.

Human hair recovered at the scene of a crime or mass disaster is commonly degraded or in an advanced stage of growth. In these circumstances, nuclear DNA analysis is often unsuccessful. For these types of hair samples, microscopical and mitochondrial DNA analyses are most commonly used. Independently, these two analyses offer lower levels of discrimination than nuclear DNA analysis as neither can individualize hair samples in an open-set population. The field of biometrics has shown potential to expand upon and improve the results of methods currently used in the forensic community. Image analysis, in particular, can be used concurrently with microscopical examinations; however, no studies have been performed to specifically quantify the reliability and joint use of microscopical, image, and mitochondrial DNA analyses.

In this study, head hairs were collected from 120 individuals. These hairs were microscopically examined and images of the root, shaft, and tip were obtained. Twenty of the 120 individuals represented five maternal relative groups. Four different microscope and camera units were used to represent the variety of systems that could be present in a forensic laboratory. Image processing methods were applied to the raw images captured by each of these units. Following image analysis, the hair samples were subjected to mitochondrial DNA analysis. These methods were used individually and in combination to individualize hair samples from the sample pool. Both success and failure rates for individualization were determined.

The goal behind using image analysis was to improve and automate current methods of examining textural and structural characteristics of human hair. This was achieved by using a set of computer vision techniques. These techniques allowed characteristics in the structure, pattern, and pigmentation of human hair to be extracted and utilized under well-defined data collection and experimental protocols. Image quality assessment methods were applied to raw hair images to improve image quality before further processing. In the proposed approach, it was necessary to create a montage of different focal planes for the human hair samples. This was due to the image acquisition process, which used the extended focus method. By utilizing image-processing techniques, common challenges, such as air bubbles, could be circumvented. After segmentation and normalization of the sample, a protocol for recognition and matching of hair structures, patches, and pigmentation was developed. The protocol resulted in the individualization of the different maternal relative groups as well as individuals. In conclusion, the results of this study demonstrated that the integration of the two analyses can add objectivity to comparative examinations between hair samples.

Hair, Image Analysis, Mitochondrial DNA

A57 Preparation of a Standard for DNA Repair Reactions

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After attending this presentation, attendees will understand the significance of using a standard for environmentally damaged DNA in experimentation of DNA repair with enzymes and enhancers of the Polymerase Chain Reaction (PCR).

This presentation will impact the forensic science community by providing a quality assurance control for experiments to restore dropped-out STR alleles from environmentally damaged DNA.

Traditionally, to provide surrogates for damaged DNA, researchers have used DNase I digestion to produce fragmented DNA or UVC-irradiation (100-280nm; found only in outer space and in the laboratory in hoods and crosslinkers with germicidal lamps) to induce lesions. In this presentation, attendees will be introduced to a novel approach in which DNA is artificially damaged during simultaneous exposure to UVA irradiation (315-400nm; in sunlight that reaches earth), heat, and humidity. As in environmental exposure, this approach allows these treatments to act synergistically on the DNA to produce multiple and disparate types of lesions. Using this strategy, DNA damage caused by environmental exposure is more accurately simulated than by using the traditional methods.

A standard for damaged DNA was prepared in an accelerated weathering chamber, the Q-Sun™ Xe-3 HSC Test Chamber, designed to provide conditions under which manufacturers can study the environmental durability of their products. These include, but are not limited to: adhesives and sealants, textiles and paper, food and beverages, paints and coatings, pharmaceuticals and cosmetics, and plastics and packaging. The chamber can provide accelerated lab testing and accommodate a variety of samples like swatches, specimens of different shapes, and bottles. Variations of temperature, humidity, sunlight, darkness, and light rainfall can be programmed by the instrument's software. The chamber was used to study DNA degradation with dried, purified DNA as well as with native DNA in body fluid stains on porous or non-porous surfaces or materials.

When DNA is isolated from evidence exposed to the environment or when samples are stored a long period of time, as in cold case scenarios, the DNA is often degraded due to exposure to heat, humidity, and sunlight, if not in a shaded area. When Short Tandem Repeat (STR) analysis is performed on DNA from such samples, the outcome may be partial profiles or no profile at all. To increase the intensities of alleles that are below an assigned threshold, enhancers of the Polymerase Chain Reaction (PCR) such as Bovine Serum Albumin (BSA) or various commercial products can be added to the reaction buffer. An alternative is to use products that claim to repair the damaged DNA. In all these approaches to improve the STR profile, a standard for damaged DNA that can act as a control for repair success has been lacking. Thus, if there is no improvement in the STR profile of the DNA extracted from the environmental sample, without a control, we have no way of knowing whether the additive was active or not during the reaction.

To prepare a standard for use in STR profile enhancement or restoration, a solution of purified Raji Control DNA was dispensed on the rubber liner of a UVC-sterilized glass vial lid and exposed to UVA irradiation (irradiance: 0.21W/m², morning and late afternoon; 0.25W/m², noon) at a set humidity (about 70%), air temperature (23°C), and surface temperature (40°C, morning and late afternoon; 45°C, noon) in the environmental chamber for 24 hours. Conditions were programmed to simulate a typical summer 24-hour day in Quantico, VA, passing from morning to noon (5.5hrs), to early afternoon (4hrs), and to late afternoon/evening (5.5hrs) exposure levels with a night period (9 hrs). Following exposure, the samples were recovered in water, quantified, and

divided into aliquots for storage. STRs were measured using the AmpFISTR® Identifier PCR Amplification Kit (Applied Biosystems). The degraded DNA standard prepared as described above (n=4 independent preparations) had a partial AmpFISTR® Identifier® STR profile (with dropouts at FGA, D7S820, D18S51, D2S1338 and CSF1PO) and a general peak height reduction at several loci that could be restored using the New England Biolabs PreCR™ Repair Mix (n=24 independent reactions).

DNA Repair, Degraded DNA Standard, STRs

A58 Effective Genotyping Strategy of Forensic Short Tandem Repeat Using Next Generation Sequencing

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After attending this presentation, attendees will have a better understanding of how forensic Short Tandem Repeat (STR) genotyping using Next Generation Sequencing (NGS) is carried out from single-source and even mixed samples and what points are important on NGS data analysis for interpreting mixed samples.

This presentation will impact the forensic science community by presenting NGS data analysis methods for STR profiling from single-source and mixed samples since NGS can be used as an additional forensic genotyping tool.

STR markers have been typed by Capillary Electrophoresis (CE)-based assays based on their length variation among human individuals. STR amplicons are usually prepared by multiplex PCR with fluorescence dye labeled template-specific primers. Despite rapid typing of STR markers by automation of the method it has some limitations, such as the number of STR loci to be measured simultaneously related to the number of fluorescence dyes and the maximum size of STR amplicons. Recently, NGS has been in the spotlight as an ultimate genotyping tool to overcome the limitations of CE-based STR analysis in forensics. STR profiling using NGS has become available along with the advancement of bioinformatics tools. Although NGS platforms produce shorter reads, they also produce a vastly greater number of reads than traditional Sanger sequencing. Hence, appropriate data analysis protocols may be required for STR profiling using NGS.

In this study, NGS libraries barcoded with Multiplex Identifiers (MID) were prepared after generating STR amplicons by multiplexing from two single sources and their 1:1 mixture samples. The libraries pooled in a mix were sequenced on a plate of Roche 454 GS Junior platform, which was selected for full sequencing of STR loci with long amplicon sizes (>300bp). NGS data obtained was sorted into three datasets according to the MID sequences. The following four steps were carried out to determine STR alleles: (1) design of STR reference sequences for common STR alleles; (2) alignment of NGS reads with reference sequences; (3) calculation of coverage (Number of NGS reads) mapping to alleles; and, (4) determination of STR alleles with defined coverage threshold. STR reference sequences allowed STR profiling from NGS data to be analyzed like CE-based assays with allelic ladder systems. For

building the reference sequences, known STR repeat structures were obtained from STRbase (<http://www.cstl.nist.gov/strbase/>) and flanking sequences were obtained from human reference genome GRCh37/hg19. Most STR alleles of two single-source samples could be determined by applying a coverage threshold of 20%; however, the threshold had to be lowered to 10% for assigning STR alleles from a mixed sample. Repeat structures for each STR loci were determined by simultaneously examining length and sequence variations in STR region. Furthermore, the actual mixture ratio of a mixed sample was estimated by not only analyzing coverage ratios of the assigned alleles in each STR locus, like peak height ratios in CE-based assay, but also by examining wild-type/variant ratios from observed sequence variations. Besides the number of NGS reads containing the variants, the distribution of the variants in the forward and reverse directions were used to assess the level of reliability for identifying true sequence variants in the STR region. In addition, the male/female mixture ratio was obtained by analyzing the coverage ratio of the amelogenin locus.

In conclusion, the NGS data analysis method presented from this study enabled successful mixture ratio estimation, as well as effective STR allele calling. Therefore, this approach will be helpful to interpret and analyze STR profiles from single-source and even mixed samples for forensic investigation using an NGS system.

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Short Tandem Repeat, Next Generation Sequencing, Mixed Samples

A59 Internal Validation of the Promega PowerPlex® Fusion System Using the Applied Biosystems® 3130xl Genetic Analyzer

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After attending this presentation, attendees will learn about Promega's new STR multiplex kit PowerPlex® Fusion and the internal validation process undertaken at the Marshall University Forensic Science Center to ensure functionality of the kit. The PowerPlex® Fusion System provides a larger number of loci than previous Promega® STR kits which results in a higher discrimination level with forensic casework and paternity samples.

This presentation will impact the forensic science community by introducing studies performed to internally validate the PowerPlex® Fusion System as well as highlighting other characteristics of the kit.

Prior to placing a new method into service in a crime laboratory setting, accredited laboratories must perform internal validations according to Standard 8 of the Federal Bureau of Investigation (FBI) Quality Assurance Standards to verify that developmentally validated methods work reliably and robustly. An internal validation was performed at the Marshall University Forensic Science Center DNA Laboratory on the PowerPlex® Fusion System amplification kit in conjunction with the Applied Biosystems® 3130xl Genetic Analyzer. This kit is a 24-locus multiplex used for human identification applications. Autosomal STR loci include the 13 core

CODIS (U.S.A.) loci as well as the 12 European Standard Set (ESS) core loci in addition to D2S1338, D19S433, Penta D, and Penta E. The kit also includes the sex-determining Amelogenin locus and the Y-STR DYS391 to identify null Y alleles from Amelogenin. The kit utilizes a 5-dye chemistry and is able to function with most genetic analyzers including the ABI PRISM® 310 and 3100 as well as the Applied Biosystems® 3130, 3130xl, 3500, and 3500xl.

The sensitivity study sought to identify optimal values for template load (15pg-10ng), PCR cycle number (29, 30, and 31 cycles), and injection time (3, 5, 10, and 15 seconds). Data from extraction negative controls were used to calculate analytical thresholds per dye channel. Genotype results of 35 samples previously analyzed using PowerPlex® 16 were compared to PowerPlex® Fusion genotype results to check for concordance. Precision was assessed by calculating the standard deviation of base-pair sizes assigned to peaks from multiple replicates of allelic ladder (n = 16 injections) and an in-house National Institute of Standards and Technology (NIST) traceable sample (n = 24 amplifications/injections). The threshold chosen for acceptable precision was three times the standard deviation and must be less than 0.5 bp. Mixture characteristics were studied by creating two-person mixtures (male-male and female-male) at ratios of 1:1, 4:1, 9:1, and 19:1. In addition, a mixture of three males at a 1:1:1 ratio was analyzed. Data from single-source samples (n = 35) were analyzed to calculate stutter formation percentages. Heterozygote peak height ratios were calculated using the sensitivity study amplification samples as well as 35 convicted offenders' samples. Negative controls from extraction, amplification, and capillary electrophoresis setup were examined to assess the risk for contamination.

Optimal parameters for amplification and capillary electrophoresis were determined to be 0.5ng to 1.0ng template input, with 30 cycles of PCR, and injection for 5 seconds. Analytical thresholds were variable between dye channels with 35rfu for the blue channel, 40rfu for green, 55rfu for yellow, and 70rfu for red. All 35 samples previously typed with PowerPlex® 16 were concordant with PowerPlex® Fusion typing results at the loci common to both kits. Of all peaks evaluated for the precision study, the largest 3X standard deviation of base-pair sizes was 0.39bp, which falls within acceptable limits. Calculated mixture proportions obtained from electrophoresis data were generally comparable to the known donor-ratios of mixed samples. Contamination risk is low, with the note that lab personnel must exercise care when setting up laboratory procedures due to the high sensitivity of the kit. Finally, data regarding stutter formation and heterozygote balance have been evaluated to develop Forensic Interpretation Guidelines as part of the incorporation of the kit into the Marshall University Forensic Science Center's Standard Operating Procedure Manual. Future studies will include a non-human DNA study and enhancement to the mixture study including mixtures of relatives.

This project was supported by Award No. 2009-IJ-CX-K11 awarded by the National Institute of Justice, Office of Justice Programs, U.S. Department of Justice. The opinions, findings, and conclusions or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the views of the Department of Justice.

Validation, DNA, PowerPlex® Fusion System

A60 Application of Molecular Antibody Probes and Flow Cytometry to Resolve Individual Contributions from Four Person Whole Blood Mixtures and Contact Epithelial Cell Mixtures

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After attending this presentation, attendees will gain an understanding of how flow cytometry and fluorescently labeled antibody probes can be used to separate distinct cell populations within multi-person (n=4) mixtures of whole blood or epithelial cell mixtures.

This presentation will impact the forensic science community by evaluating the efficacy of immunological staining and flow cytometry analysis for processing biological evidence for DNA profiling. This method can potentially reduce interpretation errors for mixed Short Tandem Repeat (STR) profiles generated from contact epithelial cell mixtures or blood mixtures containing four or more individuals.

Mixture interpretation is one of the biggest challenges in DNA analysis. There are several techniques for analyzing mixtures with different cell types, such as epithelial cells and sperm cells. However, most of these methods cannot be used to separate mixtures containing identical cell types from different individuals. In this study, a new molecular method was developed based on variation in the Human Leukocyte Antigen (HLA) that is expressed on the cell surfaces in nearly every human tissue type. The complex of genes within the HLA loci is one of the most polymorphic gene systems known. While the discriminatory power of HLA allele types is not as high as STR analysis, the range of population frequencies for HLA alleles makes them a promising tool for differentiating individual contributors in a biological mixture. HLA-antibody labeled cells can be physically separated by Fluorescence Activated Cell Sorting (FACS) prior to DNA isolation. FACS is a type of flow cytometry analysis whereby fluorescently tagged cells are selectively isolated from a heterogeneous mixture of cells. DNA extraction and STR analysis are subsequently performed on the isolated cell population. Preliminary studies have shown that HLA probes and FACS separation can be used to analyze whole blood mixtures comprised of two individuals; however, this technique has not been demonstrated on other cell types (i.e., epithelial cell mixtures) or mixtures with more than two contributors.

In the first part of this study, four-person whole blood mixtures were tested with the HLA-probe and FACS method. Four whole blood samples were acquired from Tissue and Data Acquisition and Analysis Core (TDAAC) at Virginia Commonwealth University. HLA typing of tissue donors confirmed that only one person in the four-person mixture was positive for HLA-A02 allele. After the HLA-probe was introduced to the mixture, HLA positive and negative cell populations were clearly seen with flow cytometry. Selective binding of the antibody probe to the A02 allele was shown through the shift in fluorescence intensity for the cell population among four contributors. DNA extraction and STR profiling for sorted mixture was performed and the STR profile was compared to the single-source STR profiles of each contributor. The STR profile of the HLA-labeled mixture was single source and identical to the known reference profile at all seven loci (D3S1358, D16S539, TH01, TPOX, CSF1PO, D7S820, and Amelogenin) for the HLA-positive contributor; however, some peaks were below the detection threshold. No evidence of contamination from the other three contributors in the mixture was detected.

In the second part of this study, HLA-probe/FACS sorting was tested with epithelial cell mixtures. Epithelial cells from two individuals with different HLA types (A02+ and A02-) were used to

create the mixture. Initial HLA-probes assays performed on the 1:1 mixture showed differential binding and FACS successfully separated labeled cell populations. Three replicates were performed for each experiment. STR profiling showed that the labeled cell population was consistent with the HLA-A02+ contributor across all seven loci (D3S1358, D16S539, TH01, TPOX, CSF1PO, D7S820, and Amelogenin). Preliminary studies also indicate that different ratios of cells from each contributor impact functionality of the assay, such that the unique profile of the minor contributor is not always recovered. Future studies will explore this issue and further evaluate ratios of two-person epithelial cell mixtures to determine if cell type is the variable which impacts functionality of the assay.

The results suggest that labeled antibody probes coupled to flow cytometry protocol is a useful strategy for physically separating four individuals from whole blood mixtures or epithelial cell mixtures, non-destructively, before tPCR amplification. This approach could lead to shorter data analysis times for forensic DNA analysts, particularly for complex epithelial and blood mixtures of more than two individuals.

Flow Cytometry, HLA, Mixtures

A61 Assessing TrueAllele® Genotype Identification on DNA Mixtures Containing up to Five Unknown Contributors

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After attending this presentation, attendees will understand the applicability and limitations of genotype modeling solutions for DNA mixture problems. This presentation will show the conditions under which genotype modeling can compute a reliable DNA identification.

This presentation will impact the forensic science community by establishing the generality of genotype modeling for DNA interpretation and validating the use of Cybergenetics TrueAllele® Casework system on mixtures having many unknown contributors.

Manual review of complex DNA evidence does not fully elicit all the data's identification information. Therefore, computer methods have been developed for mathematical interpretation of mixed and low-template DNA. The genotype modeling approach computationally separates out the contributors to a mixture, with uncertainty represented through probability. Comparison of a contributor genotype to another genotype, relative to a population, calculates a Likelihood Ratio (LR). Validating an interpretation method on a broad range of DNA mixtures having known composition can help predict an expected LR outcome in a particular case.

This randomized experimental design examined 40 DNA mixture items. The four mixture sets had two, three, four, or five contributors, with each item specified as a random mixture weighting of randomly assigned known references. Both normal (1ng) and low (200pg) template amounts were studied for a total of eight groups (four contributor numbers x two template amounts) each having ten mixture items.

The Mixture Weight (MW) of each item's contributors had a predetermined design value, but was subject to laboratory variation. For each item, the TrueAllele system computed two MW

estimates, one using all the known genotypes, and the other with all genotypes unknown. MW was also computed manually on the two contributor items. There was a strong association ($r^2 = 0.999$) between the three computed MWs for an item, and less ($r^2 = 0.907$) with the design value ($p < 10^{-12}$). The computed TrueAllele known-genotype MWs had the most precise values (average $sd = 0.0195$ log(LR) units) and were used in the remainder of the study.

Following a procedure used in a previous validation study, scatterplots were developed comparing a contributor's known DNA quantity (logarithm of MW x total DNA, x-axis) versus its identification information (log of LR, y-axis).¹ This approach permitted examination of all the match results (all contributors of all items) within their groups across a single statistical analysis. The scatterplots of positive match results were roughly linear ($r^2 = 0.638$), showing expected log(LR) reductions for equal MWs and high DNA amounts. The average regression slope was 12.66 log(LR)/log(DNA) ($p < 10^{-40}$), so a ten-fold change in DNA amount yielded a trillion-fold change in LR.

Analysis of Covariance (ANCOVA) of the eight groups showed different x-intercept values, but no significant difference in slope ($p = 0.348 > 0.05$). This slope invariance was observed across four different contributor numbers (2, 3, 4, and 5) and DNA template amounts (200pg and 1ng). This invariance indicates that TrueAllele's information response to DNA mixture data is relatively independent of contributor number or template amount. The ANCOVA outcome suggests that this genotype modeling method produces reliable match results, regardless of the DNA mixture composition.

The false exclusion rate (Type II error) was estimated as a function of MW. For normal DNA amounts, there were positive match results in 100% of comparisons ($0.10 \leq MW \leq 1.00$), 82% ($0.05 \leq MW \leq 0.10$), 40% ($0.01 \leq MW \leq 0.05$), and none below 0.01. With low-template DNA, positive match results were found in 100% of comparisons ($0.25 \leq MW \leq 1.00$), 91% ($0.10 \leq MW \leq 0.25$), 24% ($0.05 \leq MW \leq 0.10$) and none below 0.05. In addition to these sensitivity and specificity results, reproducibility was measured in all groups.

This validation study used randomly generated DNA mixtures (reflective of actual casework samples) of up to five contributors, with both high and low template amounts to assess TrueAllele genotype modeling. The study found that the computer's MW values were reliable and that match information changed with DNA quantity in a predictable way that did not significantly depend on contributor number or template amount. Type II error was determined as a function of MW. This in-depth experimental study and statistical analysis show the applicability and limitations of the TrueAllele method.

Reference:

1. Perlin, M.W. and Sinelnikov, A. An information gap in DNA evidence interpretation. *PLoS ONE*, 4(12):e8327, 2009.

DNA Mixture, Genotype Modeling, Validation Study

A62 Cross-Laboratory Comparison of Next Generation Sequencing (NGS) Instrumentation and Bioinformatics Software Tools for Forensic Mitochondrial DNA Analysis

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After attending this presentation, attendees will have a better understanding of the current efforts of community members to assess and validate Next Generation Sequencing (NGS) technologies for human mtDNA analysis in forensic science. Focus will be placed on analysis of data generated with three benchtop sequencing instruments using multiple commercially available software packages.

This presentation will impact the forensic science community by describing inter-laboratory studies designed to evaluate sequencing methods that will ultimately be implemented in crime laboratories in the future.

Next-generation sequencing methods are proving to be particularly well-suited for mitochondrial DNA analysis and may provide forensic analysts with a powerful tool that enables deconvolution of mtDNA mixtures or accurate quantitation of low-level heteroplasmy. Recently, Illumina® has been working with members of the community to establish a human mtDNA Forensic Genomics Consortium (IFGC) for concerted evaluation of NGS methods for potential use in mtDNA casework and databasing. In June 2013, a set of samples was prepared consisting of quantified buccal extracts from two donors as well as a series of mixtures of the buccal extracts at defined ratios (5, 2, 1, and 0.5%). Initially, whole mitochondrial genome reference data was generated at The Armed Forces DNA Identification Laboratory (AFDIL) for comparison purposes using Sanger methods. The sample set was also distributed to other participating IFGC laboratories for sequencing on multiple benchtop NGS platforms including the Ion Torrent Personal Genome Machine (PGM™), Roche GS Junior, and Illumina® MiSeq, to enable an inter-laboratory comparison of sequencing methods using identical samples. At the National Institute of Standards and Technology (NIST), the mtDNA genome was amplified from each sample set using a three amplicon long-PCR strategy, and NGS libraries were prepared using methods defined by Life Technologies™. Prepared libraries were then sequenced on the PGM™. In the laboratory, the samples were sequenced on both the Roche GS Junior and Illumina® MiSeq NGS platforms. Libraries from hypervariable regions were sequenced on the Roche GS Junior using an amplicon library preparation approach where PCR primers were designed to include required adaptors and multiplexing indices. For sequencing on the Illumina® MiSeq, libraries were prepared using Nextera® XT in which two large amplicons covering the whole mtGenome, as well as HV1 and HV2 amplicons, were enzymatically fragmented, and adapters and indices incorporated during a limited-cycle PCR reaction. The data generated by all three platforms was analyzed using SoftGenetics® NextGENe®, CLC Genomics Workbench, and a new mtDNA Variant Caller package designed by Illumina®. Sample dependent variant calls generated with each software package were compared. Data analysis with all third-party software packages resulted in detection of all expected variants in single-source samples. In mixed samples, positions expected to contain variants were evaluated for variant call present, coverage depth, and concordance with the sample associated mixture ratio. Bioinformatic processing of data using alignment algorithms specific to each software package gave rise to differences in indel alignments. These differences will easily be eliminated when an appropriate alignment algorithm is identified for analysis of all forensic mtDNA NGS data. Additionally, sequence pile-ups showed evidence of sequencing errors associated with homopolymeric regions. However, data from the MiSeq showed very low background noise overall, especially in these homopolymeric regions. Further, different third-party data analysis software packages yielded similar data for all samples, with some exceptions. Additional research is needed to further assess differences that arise as a result of bioinformatic approaches used for data analysis and the effects of these differences on

interpretation. The Illumina® MiSeq offers a streamlined enzymatic library preparation approach, low background noise overall, and high-throughput. As a result, the Illumina® MiSeq is well-suited for forensic mtDNA analysis in both casework and databasing laboratories.

mtDNA, Next Generation Sequencing, Bioinformatics

A63 Validation of a Mitochondrial DNA Testing Kit for Forensic Casework

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After attending this presentation, attendees will understand the advantages of a commercially available kit for mitochondrial DNA testing and the quality control testing that is required for its design and delivery.

This presentation will impact the forensic science community by providing a scientific basis for a commercial mitochondrial DNA amplification kit that is optimized and subjected to quality control testing with the aim of increased efficiency. All of this will decrease the amount of time the forensic DNA analyst must take to order primers and reagents, combine reagents, and perform the laboratory work.

There are dozens of laboratories world-wide evaluating the Control Region of the mitochondrial genome for forensic casework and research purposes. These laboratories process thousands of samples each year to include reference standards, hair, bone, teeth, nails, blood, and tissue. Interestingly, there is no commercially available kit simply for the amplification and resultant product for downstream Sanger dideoxy sequencing. A mitochondrial DNA (mtDNA) kit similar to the autosomal and Y-STR kits, commercially available for forensic DNA testing and databasing or clinical molecular assays available for patient testing and research purposes, would streamline laboratory processing and increase the efficiency of mtDNA testing laboratories.

Mitochondrial DNA sequencing is a laborious process. In forensic mtDNA testing, samples are extracted and two regions of the displacement loop (D-loop) are amplified: hypervariable region 1 (HV1) and hypervariable region 2 (HV2). This process includes the following steps: DNA extraction, DNA quantification and normalization, HV1 and HV2 amplification, cycle sequencing, electrophoresis, and analysis. Presented is a new mtDNA amplification kit. This kit contains the primers (HPLC-purified and pre-aliquotted in 100 nmole vials), a master mix, and control DNA. The master mix contains all the necessary components for amplification to include a buffer, an enzyme, CleanAmp™ dNTPs, magnesium chloride, bovine serum albumin, and water. Reducing the number of reagents the analyst is required to add to the amplification reaction decreases human error and the overall preparation time. In addition, Quality Control (QC) testing of each of the reagents is already conducted.

Developmental and internal validation studies were performed to include sensitivity studies, stability studies, and reproducibility studies. A total of 15 buccal swabs and 15 bloodstains were collected using different storage and collection media. In addition, extracted DNA from 20 adjudicated cases was obtained. These 50 samples were successfully amplified using the mtDNA

amplification kit. The amplified product was purified using ExoSAP-IT®, cycle sequenced with the “dilute-and-go” off-the-shelf primers from the kit and BigDye® Terminator v1.1, and unincorporated dye terminators purified with the BigDye® XTerminator™ Purification Kit. Then, they were subjected to electrophoresis on the 3130xl Genetic Analyzer and the sequence data were analyzed using eFAST™ Software v2.0 and MTextpert™ Software. All sequence data produced medium- to high-quality trace scores on Sequence Scanner Software v1.0.

This mtDNA testing kit greatly enhances the throughput capabilities of mtDNA testing for forensic and research laboratories. Reducing the number of preparation steps and sample transfers allows for the streamlining of the process and creates a procedure highly amenable to robotic adaptation. Effectively decreasing the time and human error for processing a sample leads to greater efficiency for the laboratory.

mtDNA, QC Testing, Validation

A64 Clothing Examination: Evidence of Exposure to Heat

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After attending this presentation, attendees will be aware of microscopically observable evidence of exposure to heat, including thermal damage, to garments and deposits of heat-damaged materials transferred from other items.

This presentation will impact the forensic science community by explaining how findings of thermally altered threads, fabric fibers, zippers, and trim materials can provide evidence of exposure to heat, whether on the sleeves of an arsonist standing by a fire at its onset or fiber melt around a bullet entrance hole, and is useful in answering case questions and in reconstruction of events.

Effects of heat exposure can be found on clothing items as either thermal alteration to the clothing itself or deposits of other heat-altered materials. Is a stain, deposit, discoloration, or fabric rupture heat damage or something else? Is it from an altered substrate or transfer from another thermally altered source? Thermal alteration also occurs in trim, buttons, stitching threads including on buttons, zippers, silk-screened designs, and nametags. A set of clothing often includes fabric of several types, as well as leather and synthetic imitation leather. The focus of this presentation is the thermal damage and alteration observable on clothing during examination with a stereomicroscope.

Heat damage to materials is encountered not only in cases involving fires, but also in shooting cases, vehicle-pedestrian collisions, and civil cases involving accidental or negligent fires.

A bullet strikes a plastic zipper causing melt damage and rupture of the margin of the underlying tightly woven nylon windbreaker fabric and penetrates the shirt beneath. Despite an absence of gunpowder particles on the jacket, the fiber melt suggests that the shot was fired from a relatively close range.

A man watching a house burn across the street told police he was just passing by. The officer, noting a smell of gasoline, submitted his shirt to the laboratory. The front of the wool shirt exhibited curled and swollen heat-altered fibers and partly melted thread securing one of the buttons.

The blue shirt of a man who had been run over was greasy and crumpled on the shoulder. A suspect vehicle was found several weeks later. When the undercarriage was examined, a green acetate fiber was found on the tailpipe. Testing demonstrated that the shirt fibers turned green upon sustained exposure to heat.

A very large man wielding a bloody knife and wearing only pants and a padded jacket was shot by police after cutting his wrists and failing to heed a command to stop. How many shots were fired? From which direction? The officer said three shots, but a number of wounds were found in the body and even more defects were found in the jacket. The man survived, so the bullet path in the body could not be explored as it would in autopsy. Two apparent bullet holes in the jacket exhibited fiber melt at the margins, establishing bullet entrance, and lead deposits were detected around a third. The bullet path through folds in the jacket was reconstructed by examining damage margins. Three shots were fired as the officer said, but from a different angle.

To investigate such scenarios, a rough profile of heat damage to different fabric types needed to be established and exemplars were prepared on garments and swatches of different fabrics: cotton denim blue jeans, corduroy slacks, coated cotton Capri pants, acrylic knit jogging pants, polyester trousers, polyester blend trousers, and silk pajamas. In addition, the same types of damage were produced on a nylon jacket shell, a loose-weave acrylic sweater-blouse, and on samples of heavy polyester twill, wool, and rayon fabrics. Each fabric was exposed to heat from a match flame and from contact with the hot burner of a stove. Examinations performed with stereobinocular microscopes at magnifications in the range of 6x to 50x.

In the polyester, nylon, and silk fabrics, light damage included fabric discoloration and light singeing. Higher-temperature exposure produced melting with decomposition resulting in bubbly crusts.

In the silk, rayon, and acrylic sweater fabrics exposed to brief higher-temperatures, the ends of the raised surface fibers exhibited tiny dark melt beads at the margins of the heated area.

Thermal decomposition, but no melting, was observed in the acrylic jogging pants, resulting in brown bubbly crusts and fiber contraction and swelling.

Cotton fabrics exhibited light charring and singeing that was difficult to distinguish from soot or fine soil deposits; heavier charring was accompanied by sticky amber residue. When touched to the hot burner, the cotton Capri pants exhibited similar surface deposits and a clear hard circular plastic deposit. The underside of the deposit exhibited a plastic film, slightly browned, peeling away from the center, revealing a thin plastic coating that melted, seeped into the weave, then solidified on the opposite side.

Thermal Damage, Clothing Examination, Reconstruction of Events

A65 Evaluating the Specificity of Genotypic Inference With TrueAllele® Casework Software

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After attending this presentation, attendees will understand how probabilistic genotyping software can reliably be applied to low and mixed template samples and how the method provides DNA match information that is both reproducible and highly specific to known donor profiles.

This presentation will affect the forensic science community by showing how computer-based quantitative data modeling removes the subjectivity common to inclusion/exclusion methods, uses more of the available genetic information, and ultimately expands the scope of suitable evidentiary samples by providing weight of evidence to profiles that are currently deemed inconclusive.

The forensic literature has increasingly made

recommendations for the use of probabilistic genotyping, including most recently a strong encouragement from the DNA Commission of the International Society of Forensic Genetics (ISFG) to adopt Likelihood Ratio- (LR) based approaches that include drop-in and drop-out for solving mixed template samples.^{1,2} TrueAllele Casework is a fully continuous Bayesian method that uses an iterative Markov chain Monte Carlo (MCMC) method to infer genotypes from evidentiary profiles and compute DNA match statistics and can easily accommodate drop-in and drop-out.³ By preserving more identification information, the computer is also able to add increased specificity to genotypic inference, ultimately resulting in a high degree of separation between known donor and non-donor likelihood ratios.

This study evaluated the specificity of computer-based genotype inference for both single-source low template and two- and three-person mixed template samples. The single-source sample data included two sets of serial dilutions (3.9pg to 500pg) amplified with Identifiler® Plus at 28 cycles. MCMC run time was 50K cycles (25K burn-in and 25K read-out) with all identification requests solved in duplicate. Single known donors were run against 19 non-donor staff profiles. Pooled data show a mean separation of 31.03 log units (nonillion) between known donor and non-donor reference profiles. All non-donor reference log(LR) values were negative with a maximum value of -2.65. The data show that a lower limit of approximately 15.6pg (the equivalent of roughly five diploid cells) can be placed on the software, below which results were not accurate or reproducible.

The two-person mixtures examined (two sets in mixture ratios of 1:1, 1:2, 1:5, 1:9, and 1:19) were also run in duplicate using 50K MCMC cycles (all mixture samples compared a total of 20 reference profiles against each other). The data show a mean separation of 36.70 log units (undecillion) between known donor and non-donor reference profiles. Non-donor log(LR) values were negative for all mixtures tested, with a maximum value of -6.76. Further, all known donor log(LR) values were positive, with a minimum log(LR) of 5.87. A suite of three-person mixtures (two sets in mixture ratios of 1:1:1, 1:2:1, 1:2:3, 1:5:1, 1:10:1, 2:2:1, and 3:3:1) also run in duplicate using 100K MCMC cycles (50K burn-in and 50K read-out) show a similar pattern, with a mean separation of 26.67 log units (septillion) between known donor and non-donor reference profiles. For all three-person mixtures tested, non-donor log(LR) values were negative with a maximum value of -3.50 and all known donor log(LR) values were positive.

Results from this study show that probabilistic genotyping match statistics were both reproducible and specific to all known donor reference profiles. Consistent with previously published data, the overall information gain provided with TrueAllele Casework was also several orders of magnitude greater than the threshold-based CPI method.⁴

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Probabilistic Genotyping, Likelihood Ratios, DNA Mixtures

A66 Secondary DNA Transfer: Could Secondary DNA Transfer Erroneously Place You at a Scene of a Crime?

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After attending this presentation, attendees will appreciate the impact that secondary DNA transfer will have on the interpretation of DNA typing results.

This presentation will impact the forensic science community by demonstrating that secondary DNA transfer does occur and that it can have a significant and detrimental impact on the interpretation of DNA typing results.

The learning objectives of this research project are to evaluate the detection of secondary DNA transfer using current technology to assess the impact of secondary DNA transfer on the interpretation of DNA typing results and to investigate what type of surface, rough or smooth, might facilitate secondary DNA transfer. The hypotheses for this project are: (1) secondary DNA transfer does occur and will complicate interpretation of DNA typing results; and, (2) rough-surfaced objects will be more conducive to secondary DNA transfer than smooth-surfaced objects.

Touch DNA evidence, which is often of low quantity and low quality, has become increasingly common in forensic investigations. As the demand for touch DNA analysis has grown, the equipment and chemistries used in forensic DNA laboratories has become more sensitive to improve the likelihood of obtaining DNA profiles from these low-template and low-quality samples. The increase in sensitivity in most cases is beneficial, but can lead to the detection of extraneous DNA not related to the forensic investigation and can complicate data interpretation. Typically, the introduction of extraneous DNA is from direct handling of the object by individuals not associated with the investigation; however, it may be possible that an individual could act as an intermediary, transferring DNA from another individual he/she had previous contact with to an object or person. The results of this secondary transfer of DNA could further confound the interpretation of DNA typing results because: (1) it could falsely link someone to a crime;

and, (2) it could be a potential route of contamination. Previous research has suggested that if secondary DNA transfer does occur it will have minimal impact on data interpretation. This presentation will impact the forensic community by demonstrating that secondary DNA transfer does occur and that it can have a significant and detrimental impact on the interpretation of DNA typing results.

The research project used handshaking to simulate contact that could lead to secondary DNA transfer. Two types of knives, smooth-handled and rough-handled, were manipulated by the participants. The knife handles were subsequently sampled for DNA using a wet swabbing technique often utilized in the collection of forensic evidence. The samples were amplified with the Identifiler® Plus Amplification Kit and analyzed on an AB 3130xl (16 capillary) genetic analyzer.

The results of this study demonstrate that, under certain conditions, secondary DNA transfer can occur. However, no significant difference in DNA yield between smooth and rough surfaces was observed as determined by an independent samples t-test. DNA typing results indicated that secondary DNA transfer occurred in 15 out of 24 samples or in 63% of the samples. In four of those samples, the DNA profile of the secondary contributor was either the only DNA profile or the major DNA profile detected. If these results were presented during a trial as forensic evidence, they would be difficult to dispute. In six samples, it was no longer possible to determine the primary handler of the knife. Consequently, the presence of secondary transfer DNA compromised the interpretation of DNA typing results.

The presence of secondary DNA transfer could potentially interfere with forensic investigations. In fact, data interpretation is complicated anytime extraneous DNA is introduced into a sample. As the sensitivity of the chemistries and equipment utilized by forensic laboratories increases and as laboratories implement low copy number DNA analysis, the likelihood of secondary DNA transfer becoming an actuality also increases. Secondary DNA transfer should no longer be disregarded as an event that may only occur under optimal experimental conditions.

Secondary DNA Transfer, Interpretation of DNA Data, Touch DNA

A67 Development of a Highly Sensitive Human and Male Quantification System for Assessing DNA Quality in Forensic Samples

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After attending this presentation, attendees will understand the use and application of a quantification test method that can simultaneously estimate the quantity of human and male DNA and quality of DNA in a forensic sample.

The presentation will impact the forensic science community by providing forensic DNA analysts with information regarding a newly developed DNA quality and quantity assessment method. This highly sensitive system is a valuable tool for making decisions regarding the appropriate test kit to select when analyzing potentially degraded forensic DNA samples.

In recent years, real-time PCR has become the standard for quantifying the amount of human genomic DNA in a forensic

sample. Currently used systems include Quantifiler® Duo, Plexor® HY, and Quantiplex™ HYres. Recent advances in mini STR analysis systems have now made it possible to analyze highly compromised samples. A system which can simultaneously provide human and male quantitative information, as well as assess the extent of degradation in a forensic sample, will be a useful tool for DNA analysts. The goal of this study is to validate and determine the utility of a real-time PCR system to assess degraded total human DNA, as well as total male DNA. Recent scientific literature reports the evaluation of the quality assessment of degraded DNA samples using Ya5-lineage *Alu* genetic element; however, these assays lack in sensitivity or do not exhibit high PCR efficiencies.¹

Alu are Short Interspersed Elements (SINE), approximately 300bp insertions which are distributed throughout the human genome in a large copy number. An advantage of the *Alu* system is the presence of a large number of fixed insertions. It has been reported that only 20% of the Yb-lineage *Alu* elements are polymorphic for insertion presence or absence in the human genome.² Because a large number of these fixed elements are present in every human genome, this minimizes the individual specific variation possible when using a single copy target quantification system. The use of an internal primer to amplify a segment of an *Alu* element allows for human specificity as well as high sensitivity when compared to a single copy target. In this system, primers and TaqMan probes were designed using two independent intra *Alu* insertion targets. To assess the quality of human DNA, the system utilizes two independent genomic multi-copy targets to obtain quantification of an 80bp "short" DNA fragment and a 290bp "long" DNA fragment in a DNA sample. The 80bp target sequence is from an *Alu* insertion whereas the 290bp target sequence is from a separate retroposon. The system also uses a male target in a region of the human Y chromosome DNA containing a sequence which is deleted on the human X-chromosome in an X-Y chromosome homologous region.³ A synthetic target is used as an Internal Positive Control (IPC) and provides an additional assessment for the presence of PCR inhibitors in the test sample. The ratio of the quantity of the long target to the short target will provide an assessment of the extent of degradation in the sample.

Developmental validation studies will be discussed including sensitivity, mixture, reproducibility and precision, species specificity, and concordance studies. Inhibition and degradation studies will be presented. A correlation study of estimated quantification for both 80bp and 290bp fragments with the STR analysis results obtained from degraded DNA samples will be presented.

In conclusion, a DNA-based qualitative/quantitative/inhibition assessment system that accurately predicts the quantity of human and male DNA as well as the quality of a biological sample will be a valuable tool for deciding which DNA test kit to utilize when processing forensically compromised samples for DNA testing.

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Quantification, Degradation, *Alu*

A68 DNA Identification of a Sexual Assailant Vasectomized Male Involved in Rape Case by Using Y-Chromosome STRs

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After attending this presentation, attendees will understand how Y-chromosome Short Tandem Repeats (STRs) and male urethral epithelial cells from vaginal swabs will be helpful in rape cases where no sperm is found in the vaginal swab or penetration without ejaculation by sexual offenders occurs. Attendees will also become aware of mixture DNA samples where more than one donor was involved and Y-STR testing yielded full male profiles from the epithelial fraction of the vaginal swab.

This presentation will impact the forensic science community by describing a new technology using Y-chromosome STRs and male urethral epithelial cells from vaginal swabs. If a vasectomized person is involved in a sexual offense with complete lack of semen, how can investigators trace that person from mixed vaginal samples? Y-STRs are important forensic tools in mixture analysis largely because of their ability to target and detect male-specific DNA. This fact is very advantageous when analyzing evidentiary samples that contain high levels of female DNA and only a very small amount of male DNA. It is proposed that this technique would be useful in rape cases where no sperm were found on victim vaginal swabs, penetration without ejaculation, and more than one donor was involved in the case.

The main focus of the research is to determine the number of male contributors in multiple rape cases and to identify the parameters of sperm that are not present in female fluids, but are found in semen, such as Y chromosome markers. A number of samples taken from vaginal swabs were collected by medical practitioners at varying intervals from first day to fourth day (i.e., after the incident 24h, 48h, 72h, 96h, respectively). Additionally, a 2ml intravenous blood sample from a vasectomized person and those involved in the sexual assault case were taken, with proper penile swabbing for epithelial cells.

Using an AmpFISTR® Yfiler® Kit Identifier kit, DNA was extracted from the vaginal swab and blood by differential organic extraction and organic extraction.

The study demonstrates the presence of the male epithelial cells in the postcoital vaginal tract using a commercially available proof. Results demonstrated that the Y-chromosome was in intact epithelial cells on postcoital days one to four. Y-chromosome positive epithelial cells were identified in vaginal swabs obtained following intercourse with no ejaculation. A complete DNA profile of these vasectomized males was obtained for all 16 Y-STR loci, namely DYS392, DYS390, DYS385 a/b, DYS393, DYS3891, DYS38911, DYS391, DYS19, DYS439, and DYS438, residing on the Y-chromosome and Amelogenin. A total number of five alleles were identified in 16 loci, indicative of a typical mixture sample with wide variation in the yield extracted from DNA 0.5ng to 1,000ng. The variation was attributed to the number of epithelial and/or white blood cells that are present in these samples from vasectomized individuals.

Amplification of Y-STR loci provides critical information during analysis of male-female mixture samples as are often found in rape cases. Genotype results come from these samples where multiple Y-STR alleles were observed at more than one locus with female alleles not found 30/32.2 and male allele not found in vaginal swab 28/31.2. In loci Y_DYS_635, Y_DYS_392, and R_Y_Gata_h4, more than one copy was found on Y-chromosome and confirmed by genotyping of 16 loci. More remarkable results came from loci G_DYS385, Y_DYS635, and Y_DYS392 in which haplotype duplication was observed. In particular, rape cases after

four days with semen from a vasectomized male did not preclude the presence of Y-chromosome from male DNA, resulting either from the presence of lysed sperm cells or male urethral epithelial cells.¹

The impetus for this study suggested in sexual offenses that semen persists in the vagina or in the cervical region of the vagina for 72hrs and up to three days in victims of sexual assault. It verifies that semen does not persist longer in the vagina than spermatozoa because total cell lysis happens very quickly due to metabolic activity change in the vagina. The present study also suggests that the medical examination of female victims and collection of vaginal swabs be performed within three days.

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DNA, Sexual Assault, Y-STR

A69 Toward a Forensic Microdevice on a Rotation-Driven Platform and Integration of a "Pinwheel" Quantitation Module

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After this presentation, attendees will better understand the forensic applications of microfluidic platforms, micro Total-Analysis-Systems (μ TAS), and the challenges of integrating forensic DNA workflow onto a single microdevice.

This presentation will impact the forensic science community by describing an integrated plastic microdevice for DNA extraction and amplification for use in Short Tandem Repeat (STR) analysis. Additionally, it will describe a conceptual quantitation module for integration onto the existing microdevice. This work presents a step toward a μ TAS that eliminates expensive manufacturing and external attachments and also mimics the current workflow in forensic laboratories.

Compared to conventional methods, a μ TAS has several advantages including smaller reagent volumes, decreased sample handling and tube-to-tube transfers, and the potential to significantly decrease sample-to-answer time. Two key technologies, Infrared (IR)-mediated Polymerase Chain Reaction (PCR) and a new DNA preparation chemistry, have allowed for greater possibilities in the microfluidic analysis of DNA, especially for forensic samples. IR-PCR expedites amplification with 30 cycles completed in 30 minutes without requiring a block thermocycler. An enzyme-based DNA preparation approach offers faster reaction times, reduced sample handling, and the elimination of PCR-inhibiting reagents, while maintaining equal performance as compared with traditional extraction methods. A primary hurdle in integration is microfluidic flow control. Previous work has accomplished this with the use of active valving systems that require bulky, expensive external hardware. However, passive valves, that do not require external hardware or mechanical actuation can be employed on a rotation-driven platform using varied centrifugal force defined by rotation speed to effectively direct flow. The goal was to integrate DNA extraction/preparation and amplification by combining these technologies into a single microdevice utilizing a rotation-driven

platform.

In this study, all microdevices were fabricated in-house by laser ablation and thermal bonding of layers of Poly(Methyl Methacrylate) (PMMA), whose thicknesses were chosen to minimize the thermal mass. Modular microdevices for DNA preparation and IR-PCR were designed first, followed by a larger integrated microdevice. Microfluidic optimization was accomplished through a series of dye tests as well as the testing of various spin times and speeds on the rotation-driven platform. After establishing flow control for the integrated device, the DNA preparation module with IR-mediated heating was optimized for buccal swabs. The results of the DNA liberation and amplification were compared to the manufacturer-suggested extraction, followed by traditional PCR amplification. Previous results suggested DNA preparation, accurate metering, and IR-PCR can be achieved on a rotation-driven platform with this newly designed integrated device. Modular on-chip DNA preparation followed by conventional PCR amplification of STR loci resulted in a full STR profile concordant with a profile generated from the manufacturer's standard protocol and conventional PCR; however, challenges with this microchip design exist including the formation of bubbles between PMMA layers and the presence of surfactant in the DNA preparation buffer — both of which negatively affect fluidic control. Additionally, there are no previous studies that have attempted to incorporate the required DNA quantitation step on-chip. Optimization of bonding temperature and pressure has relieved the bubbling issue. To overcome the surfactant issues, several approaches have been taken, including increasing centrifugal speed, widening of channels, and decreasing the buffer concentration used for DNA preparation to directly reduce the amount of surfactant present. While further optimization is still required, these changes have yielded performance improvement. With each modification, microfluidic flow control is assessed and DNA yield is measured. When modifications are defined to address the surfactant and bubbling issues, these will be incorporated into the next iteration chip design and the resulting DNA from the successfully integrated chip will be tested to ensure an acceptable STR profile is generated.

Finally, it is necessary to consider DNA quantitation in order to develop a device that is consistent with the requirements of DNA Quality Assurance Standard 9.4. Thus, a fluidic module employing a "pinwheel" quantitation assay is being designed. This method measures the aggregation of silica-coated superparamagnetic beads to directly quantitate DNA. This assay does not require expensive instrumentation, is fluorescence free, and has been shown to work on a PMMA microdevice. On-chip quantitation was compared to conventional quantitative PCR; quantitation results will be discussed. Successful integration of this system, coupled with ease of fabrication, will promote progress towards a simple sample-in, answer-out microdevice that more closely mimics the current workflow in forensic DNA laboratories.

Microfluidics, Micro Total-Analysis-System, DNA Quantitation

A70 Using Secondary Ion Mass Spectrometry to Better Understand Fingerprint Aging Through Imaging and Depth Profiling

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After attending this presentation, attendees will be aware of how Secondary Ion Mass Spectrometry (SIMS) can be an important tool for studying how fingerprints are affected by both time and environmental conditions. More specifically, an understanding

of how SIMS can be used in a multi-faceted approach providing different types of analysis will be discussed.

This presentation will impact the forensic science community by providing a nontraditional method to probe the chemistry of latent fingerprints. In addition to being able to complete traditional mass spectral analysis, the community will be presented with a technique which has the ability to also depth profile a fingerprint ridge as well as obtain high-resolution chemical images. The combination of these analyses has led to better understanding of what occurs within a latent fingerprint as it ages.

Chemical analysis of fingerprints has traditionally been completed using techniques such as Infrared (IR) spectroscopy, Gas Chromatography Mass Spectrometry (GC/MS), and Liquid Chromatography Mass Spectrometry (LC/MS); however, in recent years, new techniques have begun to be applied to fingerprint analysis that allow for the specific chemical analysis of fingerprints without destroying the fingerprint. One of the techniques that is being explored is SIMS. SIMS presents several unique benefits to the analysis of a complex sample like a fingerprint, including the ability to probe both the organic and inorganic chemical composition, the ability to obtain to high resolution chemical imaging, and the ability to profile through a fingerprint ridge without damaging the ridge detail of the latent fingerprint. The goal of this study is to outline how these analyses can be exploited to understand fingerprints with respect to aging.

Initial studies have been completed with latent fingerprints that have been aged at different intervals and under different environmental conditions. These fingerprints were analyzed by SIMS using a number of procedures to obtain mass spectra, chemical images, and depth profiles. Using this data, several initial results have been uncovered, with the main focus on chemical imaging. Through initial studies, it has been shown that the location of components in a fingerprint changes as a function of the age, exposure, and surface of deposition — with a number of constituents appearing to diffuse out of the ridges of the fingerprints and into the valleys. These timescales are a matter on the order of days to months. Furthermore, it has also been found that there is a threshold after which the chemical imaging technique cannot differentiate the ridges and valleys of fingerprints because of diffusion, though the ridge detail is still optically visible. Depth profiling of the ridges and valleys of fresh and aged fingerprints has also shown that it can be used as a method to measure the depth of ridge versus the valleys. This may prove a beneficial method to age fingerprints, as the depth of the ridges decreases with time, as the depth of the valleys increases.

In addition to these initial studies, current work is looking at the reproducibility of these observations in addition to understanding the role which the deposition substrate plays. These observations, in addition to how environmental conditions (UV-exposure, ozone, heat, and humidity) affect the fingerprint, will also be discussed.

Fingerprints, SIMS, Chemical Imaging

A71 Development of Presumptive Color Tests for Emergent Drugs

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After attending this presentation, attendees will be able to crucially examine the potential of ion pairing in the color detection of emergent drugs.

This presentation will impact the forensic science community by demonstrating the potential of ion pairing to detect

emergent drugs.

Presumptive color tests for drugs have helped investigators narrow the possible identities of a substance. They are quick and inexpensive chemical tests commonly performed by police officers on the street and prior to the use of costly confirmative tests in the crime lab. These tests are done on the scene to quickly determine if the police officer has sufficient probable cause for an arrest. Using this new technique, an investigator can place the questioned substance in a disposable test tube containing ampoules of chemical reagents necessary for the presumptive identification of piperazine drugs such as Benzylpiperazine (BZP) and 3-Trifluoromethylphenyl-piperazine (TFMPP).

BZP and TFMPP are synthetic phenylpiperazine analogues which have stimulant and amphetamine-like properties.¹ As a result, they are commonly used as recreational drugs and were legally available in a number of countries, particularly in New Zealand.¹ BZP drugs were temporarily classified as a Schedule I controlled substance in the United States in 2002 because of their high abuse potential and lack of accepted medical use or safety.² On March 18, 2004, the DEA published a Final Rule in the Federal Register permanently placing BZP in Schedule I. Several states have placed BZP in Schedule I, including Iowa, Tennessee, Wyoming, Mississippi, Louisiana, Idaho, Colorado, Illinois, Indiana, Kansas, Missouri, Oklahoma, and Nebraska.²

According to the Office of Diversion Control, law enforcement officials submitted 48 drug items/exhibits to federal, state, and local forensic laboratories identified as BZP in 2004.² The number of BZP items/exhibits increased from 437 in 2007 to 6,088 in 2008. BZP items/exhibits submitted to forensic laboratories increased 127% from 6,088 in 2008 to 13,822 in 2009.

Typically, BZP is smuggled as powder through drug trafficking organizations internationally. The bulk powder is then processed into capsules and tablets. BZP is encountered as pink, white, off-white, purple, orange, tan, and mottled orange-brown tablets. These tablets bear imprints commonly seen on MDMA tablets such as a housefly, crown, heart, butterfly, smiley face, or bull's head logos and are often sold as "ecstasy." BZP has been found in powder or liquid form which is packaged in small convenience sizes and sold on the internet.² From the increasing trend in distribution and abuse cases, the presumptive test for piperazine drugs play an important role for law enforcement to identify the controlled piperazine substances within a minute on the street.

Both BZP and TFMPP appear as a colorless solution when dissolved in deionized water. In these experiments, the BZP and TFMPP aqueous solution in the presence of combinations of methyl orange and 1-(2-hydroxyl-1-naphthylazo)-2-naphthol-4-sulfonic acid zinc salt yields a yellow solution as a positive result; however, this positive result was severely interfered with by organic molecules containing primary amine functional groups. The results of the method and future plans for further improvements will be presented.

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Ion Pairing, Synthetic Cathinone, Color Test

A72 Fingernail Scrapings Collected From a Burnt Body Revealed the ID of the Suspect: A Case Study

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After attending this presentation, attendees will learn the importance of the DNA analysis on fingernail scrapings collected at the time of the autopsy. The biological material or DNA found under fingernails preserves even in extreme circumstances such as severely burned bodies. In a homicide case, the DNA recovered under fingernails from a burnt body found in a field in Cleveland, OH in a homicide case revealed the identity of the killer.

This presentation will impact the forensic science community by showing the importance of the collection and analysis of fingernails/scrapings collected at the time of the autopsy in helping to identify the perpetrator(s).

This case study demonstrates the ability of DNA to withstand extreme conditions such as fire, heat, etc. In general, fingernail scrapings or swabs collected from under fingernails of homicide victims may play an important role. In some cases this may be the only evidence that provides a link between the decedent and the perpetrator. In homicide cases involving victims burnt or charred beyond recognition, given the priority to identify the victim, it is possible that this type of evidence may be overlooked. The current study involves the homicide of a young woman whose body, charred beyond recognition, was found in the backyard of an abandoned house in Cleveland, OH.





Photo: Victim as found in the field

The Cuyahoga County Medical Examiner's office collected the evidence from the scene. Fingernail scrapings/swabs were collected from the hands of the victim at the time of the autopsy. DNA analysis was performed on the fingernail scrapings/swabs in an attempt to detect any DNA foreign to the victim. DNA extraction was performed using the Promega® DNA IQ™ Casework Pro Kit on a Maxwell® 16 instrument. DNA quantification was performed using the Applied Biosystems® Quantifiler® Duo DNA Quantification Kit on a 7500 Real-Time PCR System. The amplification was performed using the Promega® PowerPlex® 16 HS System and the Applied Biosystems® Yfiler® PCR Amplification Kit. The electrophoresis was performed on Applied Biosystems® 3130 and 310 genetic analyzers and the data was analyzed using Applied Biosystems® GeneMapper® ID software.

Male DNA was detected in DNA extracted from the fingernail scrapings/swabs from both hands of the victim. Autosomal DNA analysis of the fingernail scrapings from victim's left hand revealed only three alleles foreign to the victim. The minor autosomal Short Tandem Repeat (STR) DNA profile foreign to the victim obtained from analysis of her right hand fingernails swab was found to be consistent with the suspect. Further, the Y-STR haplotype developed from both the hands was found to be consistent with the suspect.

Fingernail Scrapings, STR DNA Analysis, Y-STR DNA Analysis

A73 Determination of Reference Sample Genotypes for the HLA-DQA1 (HLA-DQa) Locus Using Direct DNA Sequencing of PCR Amplicons

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After attending this presentation, attendees will understand the basis for an inexpensive way to genotype the HLA-DQA1 locus using the Sanger method of DNA sequencing of Polymerase Chain Reaction (PCR) amplicons.

This presentation will impact the forensic science community by showing how the direct sequencing of the PCR amplicons will allow a number of forensic biology laboratories to genotype for the HLA-DQA1 locus for the agencies they service. This may help investigators exclude or include new cold case suspects by comparing their types to previous results for unsolved cases.

The HLA-DQA1 locus is part of a human leukocyte antigen gene that is located on human chromosome 6. The locus has been

studied extensively in the past, including the determination of the allele sequences and their frequencies in various populations. The locus exhibits normal autosomal inheritance. While the locus does amplify for non-human primate species, more distantly-related animal species and bacteria do not produce amplification products.

HLA-DQA1 (HLA-DQa) was one of the first genetic loci to be amplified using the PCR for forensic casework purposes. The most recent version of the HLA-DQA1 PCR kit was commercially supplied until approximately 2002. This kit utilized a reverse dot blot typing method by hybridization of PCR amplified products onto nylon strips. The strips were spotted with attached Allele Specific Oligonucleotide (ASO) probes that detected denatured amplified products that contained a biotinylated primer. Following a color development that resulted in blue circular spots on the white nylon membrane where hybridization occurred, the sample genotype was indicated. The latest produced kit detected seven alleles for a total of 28 genotypes. Although there is still a small demand for HLA-DQA1 testing, the reverse dot blot kits from Perkin Elmer® are no longer available. The PCR kits last produced expired in 2003.

An increasing number of law enforcement agencies are creating and participating in "cold case" investigations and capital case reviews are also being conducted by appellate attorneys. In a number of these cases, the evidence has been discarded, lost, or was exhausted in the original analysis. What remains, in most cases analyzed in the 1990s, is a report and/or notes with at least HLA-DQA1 results from the evidentiary samples in the case. After review and investigation by cold case detectives, quite often a new suspect is developed. When this occurs, this suspect will need to be compared to the forensic DNA typing results obtained during the original case analysis.

While the previous method used a hybridization procedure, this presentation describes a procedure for the direct sequencing of the seven DQA1 alleles used in forensic analysis and the characterization of the 28 possible genotypes of the HLA-DQA1 locus for comparison to types developed during previous casework analysis. A direct sequencing method has been developed to characterize all 28 genotypes. This procedure is intended to be used primarily for typing of newly obtained reference samples with adequate DNA to allow comparison of the resulting genotypes to results of analyses performed using the former reverse dot blot kits. This procedure is not intended to be used on mixtures.

HLA-DQA1, Amplicon Sequencing, Reference Samples

A74 NIST Inter-Laboratory Studies for DNA Mixture Interpretation

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After attending this presentation, attendees will better understand the history and lessons learned from NIST inter-laboratory studies on mixture interpretation, including the most recent study in 2013 (MIX13).

This presentation will impact the forensic science community by helping the forensic DNA community to: (1) determine the current "lay of the land" regarding STR mixture interpretation across the United States; and, (2) learn where future training and research could help improve mixture interpretation and reporting.

DNA mixtures of two or more individuals can be challenging to interpret for the forensic DNA scientist. Historically, laboratories have developed protocols to interpret mixtures based upon a combination of multiple areas of investigation including (to

name a few): internal validation studies; information present in the scientific literature, training from workshops and scientific meetings; and, guidelines as reported by scientific working groups such as Scientific Working Group on DNA Analysis Methods (SWGDM) or the DNA Commission of the International Society of Forensic Genetics.

Occasionally, the laboratory may find it useful to compare their analytical results using their mixture interpretation protocols to other peer laboratories via interlaboratory studies. Since 1997, the National Institute of Standards and Technology (NIST) has conducted five DNA mixture interlaboratory studies, including: Mixed Stain Study one (1997); Mixed Stain Study two (1999); Mixed Stain Study three (2003); MIX05 (2005); and, MIX13 (2013). The first three mixed stain studies involved a "wet lab" design where the lab would extract and amplify the mock mixtures to generate data for analysis.¹⁻³ In general, differences between laboratories were observed due to variation in extraction efficiency, variation in DNA quantification, and variation in establishing an analytical threshold.

In MIX05, NIST focused on electronic data for analysis to control for the variation observed in previous studies with extraction and quantification.⁴ One observation in the analysis of the results was the wide range of approaches to interpreting the same data among the different laboratories. With the publication of the 2010 SWGDAM Autosomal STR Interpretation Guidelines, many laboratories have established analytical and stochastic thresholds for mixture interpretation.⁵

The MIX13 interlaboratory study was conducted to determine the current "lay of the land" in regard to STR mixture interpretation across the community. Another goal was to gauge the consistency in mixture interpretation across the United States after the publication of the 2010 SWGDAM guidelines. Examples of the MIX13 interlaboratory study will be shared along with ideas for future training and research to improve mixture interpretation and reporting in the United States.

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Forensic DNA, DNA Mixtures, Mixture Interpretation

A75 Implementation of an Optimized Extraction Method and a Modified Amplification Protocol for Improved Y-STR Success With Compromised Skeletal Elements

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After attending this presentation, attendees will learn about modified extraction and amplification strategies to improve the recovery of nuclear data from decades-old skeletal specimens.

This presentation will impact the forensic science community by demonstrating improved forensic DNA methods can obtain additional genetic information that can assist with the re-association and identification of commingled remains in missing persons cases.

The identification of compromised skeletal remains at the Armed Forces DNA Identification Laboratory (AFDIL) is primarily achieved via mitochondrial DNA (mtDNA) analysis. Although mtDNA typing offers the greatest chance of success, its forensic utility is often limited by its maternal inheritance and requires the availability of appropriate references. Data from alternative DNA markers in the nuclear genome would benefit identification efforts, but the poor quality and limited quantity of nuclear DNA present in degraded samples has historically restricted the use of this data. To address these limitations, a modified amplification strategy using a commercial kit to recover Y-chromosomal Short Tandem Repeat (Y-STR) data from challenging samples has recently been validated.¹ The use of this protocol required the implementation of data analysis and interpretation guidelines to mitigate stochastic effects and ensure the generation of reliable consensus profiles. Validation data has demonstrated a 45% increase in Y-STR profile recovery as compared to the standard amplification protocol. Further improvement has been observed when this method is coupled with an optimized version of the previously published demineralization protocol, which includes a purification step using silica columns and eliminates the need for organic reagents.^{2,3} Presently, a successful Y-STR profile (four or more reportable loci) can only be generated in 24% of cases processed with organic extraction and standard amplification. Preliminary results based on a limited number of cases have shown Y-STR success increases by 33% when the two modified protocols are applied in tandem to compromised skeletal elements. The implementation of these two procedures at the AFDIL promises to provide additional genetic information to assist with the re-association of commingled remains and support identifications in missing persons cases.

The opinions or assertions presented hereafter are the private views of the author(s) and should not be construed as official or as reflecting the views of the Department of Defense, its branches, the U.S. Army Medical Research and Materiel Command or the Armed Forces Medical Examiner System.

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Degraded Skeletal Remains, Missing Persons, Validation

A76 Efficient Strategies for DNA Identification: ICMP Experiences Over Time in the World's Largest Missing Persons Identification Project

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After attending this presentation, attendees will gain an understanding of factors to be considered in conducting DNA testing in large-scale identification projects, with an emphasis on cost- and time-effectiveness at various stages of a project, and the use of additional and/or newly developed DNA marker systems to find difficult DNA matches.

This presentation is intended to impact the forensic science community by contributing to a better understanding of how modern techniques of DNA testing and matching can be applied to a program of missing persons identification on a large scale, and how such projects may be devised to maximum cost effectiveness.

The International Commission on Missing Persons (ICMP) was initially established to assist with the problem of large numbers of persons missing from conflicts in the former Yugoslavia that occurred in the 1990s. Some ~40,000 missing persons were unaccounted for. Many of the missing were deposited in mass graves and recovered in a mostly degraded, and often highly commingled, skeletal state. The lack of medical and dental records for a large number of individuals, combined with the overall context, rendered "traditional" identifications generally inefficient and unreliable. In 2000, the ICMP launched a "DNA-led" blind matching program, applied regionally. In a massive outreach campaign, blood samples were obtained and genetic profiles generated from family members of the missing, and compared to DNA profiles obtained from skeletal remains in a high throughput testing system. Software-driven semi-automated DNA matching has been performed to issue a large number of DNA Match Reports with a high degree of objective surety, in support of identifications.

The primary ICMP databases of victim and family genetic profiles were established with a 15-locus STR multiplex (Promega® PowerPlex® 16), with a high success rate in obtaining useful DNA profiles from degraded skeletal remains using optimized DNA extraction protocols. Focusing on Bosnia, the ICMP DNA

database contained, as of August 2012, 32,236 bone DNA profiles representing 16,750 different individuals and 71,265 family reference DNA profiles representing 23,385 missing persons. From these, 13,988 individuals have been DNA matched at very high statistical surety, providing the basis for identification. Overall, for the former Yugoslavia, 16,670 missing persons have been DNA matched, for a matching rate of approximately five per working day over the entirety of the project.

The blind matching approach employed has been highly efficient over the years, enabling matches to be made at an unprecedentedly high rate and scale, providing answers to families as quickly as possible and as inexpensively as possible with very high rigor and surety. However, the project has now reached a stage where this rate has fallen dramatically due to the low rate of discovery of new human remains, and a very low rate of collection of new family reference samples. As the project enters a phase where the "low hanging fruit" has already been harvested, additional efforts are being applied to find as many matches as possible within the existing ~2,700 unmatched victim profiles and family reference profiles representing ~9,000 unidentified missing persons.

Results from a coordinated effort for detecting and reporting additional DNA matches will be presented, based on the following additional approaches: (1) intensified screening of databases for possible family associations with low evidentiary support at present; (2) confirming or refuting possible family associations through the application of additional testing systems; and, (3) re-visiting samples that previously failed to give profiles using newer highly optimized extraction methods, "mini-STR" kits, and additional purification steps. Additional STR multiplexes have been validated to provide 23 loci, and application of these additional markers has resolved many cases, with an average increase of likelihood ratio on the order of 10^4 (in cases that are not exclusions with the new markers). Likewise, selected cases are now being queried with mtDNA and Y-chromosomal testing.

The results of these efforts will be summarized and discussed from the standpoint of cost-benefit ratio at various stages of a massive identification project. Strategic and policy implications for the very long term will also be discussed in the context of this and other missing persons projects whose scale and complexity will pose issues for many years.

Missing Persons, DNA Identification, Family Matching

A77 OSIRIS Version 2.2: Intelligent Software for Casework and CODIS DNA Analysis

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After attending this presentation, attendees will have an understanding of the underlying principles of Open Source Independent Review and Interpretation System (OSIRIS) Short Tandem Repeat (STR) DNA profile analysis software, its basic use and capabilities, some forensic uses, and how it can be integrated into manual or automated workflows.

This presentation will impact the forensic science

community by informing it about free, open-source STR analysis and quality control software designed to increase the efficiency and accuracy of STR DNA analysis in forensic casework, convicted offender profiling, and other identity testing laboratories.

With resources limited, and the volume of forensic DNA profiling increasing, there is a need to increase DNA analysis efficiency and accuracy. OSIRIS, the Open Source Independent Review and Interpretation System, is freely available software for the analysis of STR profiles produced using Applied Biosystems® 310, 31xx, 3500, and 3700 series Genetic Analyzers, and can be downloaded at <http://www.ncbi.nlm.nih.gov/projects/SNP/osiris/>. OSIRIS analyzes both .fsa and .hid format files produced by the collection software, using an independently derived mathematically-based sizing algorithm.¹ OSIRIS supports numerous commercially available marker kits including CODIS-compliant kits as well as other kits favored by forensic, biomedical, and relationship testing laboratories. Supported kits now include GlobalFiler™, Fusion™ and PowerPlex Y23™, and OSIRIS includes sophisticated Y-STR analysis and dynamic baselining.

OSIRIS identifies peaks by iteratively fitting expected parametric data signatures to the observed data, with matches generally having correlations in excess of 0.999. Parametric peak locations are determined as time and transformed into base pair coordinates. Rather than the global Southern method, OSIRIS instead uses the correspondence between a sample's ILS and an associated allelic ladder to map the time scale of the ladder into that of the sample, fitting the most appropriate ladder to each sample. This integration of the ladder with the sample permits a straightforward and accurate comparison of sample peaks with ladder locus peaks. This method provides two additional peak quality measures: fit level and sizing residual (a measure of peak shift).

OSIRIS is designed to be flexible for ease of integration with the laboratory's own workflow, including its Laboratory Information Management System (LIMS) and other software, reducing manual intervention in both analysis and data transfer. It increases the efficiency and accuracy of analysis by categorizing artifacts either as critical, requiring human review, or non-critical, dependent on user-designated parameters. Analysis is fast, typically under 30 seconds per 96 well plate. For poor quality samples, OSIRIS can intelligently predict required re-work conditions. An audit trail of editing by analysts and reviewers is preserved, and laboratory review procedures can be software enforced. Reporting and data export are very flexible, allowing laboratories to export tables, CODIS compatible data, LIMS-formatted data, and other formats of the laboratory's own design, and can be automated to export subsets of data directly upon analysis. OSIRIS also has quality metrics that allow it to be used as a lab process quality assurance tool.

OSIRIS version 2.2, developed in collaboration with the U.S. Army Criminal Investigation Laboratory and the Illinois State Police DNA Indexing Laboratory, specifically addresses casework analysis, low-level DNA analysis, and editing efficiency. OSIRIS v. 2.2 decreases the amount of analyst editing of artifacts and improves low-threshold analysis using dynamic baselining in the identification of low-level peaks and artifacts.

In addition to OSIRIS having been validated for forensic casework and as an NDIS-approved expert system for CODIS testing, it has been validated for clinical testing of bone marrow engraftment, as an expert system for relationship testing, and for research cell line verification in laboratories in the U.S. and internationally. Additionally, OSIRIS is used for STR analysis training and data review.

Work at NCBI is supported by the Intramural Research Program of the NIH and the National Library of Medicine.

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STR, DNA, Analysis

A78 Validation and Implementation of OSIRIS for Forensic DNA Casework and Incorporation Into Laboratory Work Flow

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After attending this presentation, attendees will be familiar with Open Source Independent Review and Interpretation System (OSIRIS), a new software program to analyze and view electropherogram DNA profile data. OSIRIS is a free, open-source, powerful analysis tool that analyzes data from instruments such as the 310, 3100/3130, and 3500/3700 and makes allele calls and artifact identification in a manner that requires less input from the scientist than traditional methods.

This presentation will impact the forensic science community by serving as an introduction to a new tool that can be used in DNA casework. This new tool is available in the public domain and has protocols for any current common Short Tandem Repeat (STR) kit. OSIRIS is also flexible enough to handle new STR kits that will be coming in the near future.

OSIRIS is a public domain software package developed by National Institute of Health (NIH) for the analysis of .fsa and .hid files generated by the forensic DNA analysis process. The United States Army Criminal Investigation Laboratory (USACIL) decided to investigate OSIRIS as a way to perform the electronic data analysis of forensic DNA casework samples. OSIRIS searches for peaks in an iterative fashion by fitting expected parametric data signatures to the observed data. Unlike traditional sizing methods for STR fragment analysis, OSIRIS does not use Southern methods to compare a sample to the ILS for base pair estimates. Instead, OSIRIS compares the ILS of a sample to the ILS of a ladder, and then does a direct comparison of the center of sample peaks to those of the ladder.

OSIRIS incorporates numerous artifact elimination strategies to minimize the number of human edits required for casework samples. These advanced artifact elimination algorithms allow for a lower peak calling threshold than otherwise possible. This, in turn, allows for more information to be preserved from a sample, particularly in instances of lower level amplification. At the USACIL, much effort went into determining a strategy to use for setting the minimum rfu peak calling threshold to take advantage of the better artifact identification strategies in OSIRIS. This resulted in a much lower rfu peak calling threshold, which in turn allows for the use of more data in the interpretation of a sample. For example, a peak calling threshold as low as 30 or 40rfu is feasible with OSIRIS, compared to 75rfu in the past.

Of note, the USACIL developed and uses ArmedXpert software for all aspects of DNA casework other than the analysis of .fsa files. Because OSIRIS is open source, the USACIL DNA Branch set about the task of integrating OSIRIS into the ArmedXpert

software currently in use. Some advantages of doing so are: (1) the ability to work in one continuous software environment; (2) not being as limited by noise and/or artifacts; and, (3) the ability to use the OSIRIS engine in the user-friendly environment of ArmedXpert.

DNA Analysis Software, Improved Artifact Filtering, Lower RFU Analysis Threshold

A79 Evaluation of Direct PCR Amplification Using Various Swabs and Washing Reagents

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After attending this presentation, attendees will learn how to process reference samples faster using various types of swabs and reagents. The goal of this research was to generate DNA profiles from body fluids deposited on different swab substrates. Each substrate containing one body fluid was treated with purification reagents prior to amplification with direct and non-direct amplification kits. DNA profiles generated were compared for concordance within and between the substrates, and between amplification kits.

This presentation will impact the forensic science community by educating the forensic DNA analysts how to process samples faster using different swabs and washing reagents.

The following substrates were used in this project: the Pur-Wraps® sterile cotton-tipped applicator; the Pur-Wraps® Foam Swab; the MiniPax®; the PurFlock® Ultra Flocked swab; the FAB-SWAB™, the Fitzco® SPIN-EZE™ Push-off™ swab; and, the Bode SecurSwab™. A 1.2mm punch of each substrate was prepared for deposition of body fluids. Saliva and blood samples from male and female donors were deposited on each punch, with 0.5µL to 5µL of saliva or 0.1µL to 2µL of blood added onto each substrate punch. The prepared specimens containing body fluids were kept at room temperature for varying lengths of time ranging from no wait time (immediate extraction) to several weeks before processing. Each single body fluid containing punch was then washed with one of the following four solutions: PunchSolution™, SwabSolution™, Prep-n-Go™ Buffer, or ECS™ wash buffer.

The protocol for the SwabSolution™ kit was modified for the substrates in this study. Each punch was incubated for the appropriate amount of time recommended, or as deemed necessary. For the ECS™ wash buffer, a protocol similar to the one employed with FTA® Purification Reagent was used. After the washing step was complete, all specimens were amplified using various amplification kits.

Autosomal Short Tandem Repeat (STR) loci and the Amelogenin gender locus were amplified using the PowerPlex® Fusion System Kit and AmpF!STR® Identifier® Direct, Identifier® Plus, and Identifier® Amplification Kits. In addition, male body fluids were amplified using the PowerPlex® Y23 System from Promega® Corporation. Amount of reagents needed, and, if necessary, amplification parameters were varied in this study in order to detect complete autosomal and Y-STR DNA profiles from all substrates and both body fluids. Recommended reaction volumes by each manufacturer were first followed in the reactions to determine if complete profiles could be generated from the substrates after treatment with each reagent. Once this process was deemed successful, half reaction volumes were used for subsequent amplifications. Analysis of the amplified products was performed by capillary electrophoresis injection on a Applied Biosystems® 3130xl Genetic Analyzer. The generated data were analyzed using

GeneMarker® HID Software Version 2.2.0.

Autosomal and Y-STR profiles were successfully obtained from all of the swab substrates containing blood and saliva samples. It was observed that the optimal Polymerase Chain Reaction (PCR) cycle number for saliva was 26 cycles, and for blood 24 cycles, when the extracted body fluid samples were amplified with the primers from Applied Biosystems® kits. However, the PowerPlex® Fusion kit required 27 cycles for generation of complete profiles from saliva. It was determined that 0.1µL of blood and 1.0µL of saliva were the optimal volumes necessary to obtain complete DNA profiles.

Concordant profiles were obtained within and between the substrates, all amplification kits, and the reagents. Half reaction volumes yielded similar results to the full reaction volumes, and both produced complete autosomal and Y-STR profiles. The modified protocols for washing using the SwabSolution™ reagent proved to be successful in generating complete autosomal STR and Y-STR profiles. This research also indicated that SwabSolution™ and PunchSolution™ performed better and more consistently in generating complete DNA profiles from the majority of the substrates. However, this was not true for all amplification kits.

In the forensic science community, implementing the use of direct PCR amplification using various swabs and different washing reagents could have an important impact. Since swabs are frequently used to collect reference samples, having the ability to process different types of swabs and amplify the body fluids more quickly using different types of kits, would be beneficial to crime laboratories as well as paternity testing laboratories and data banking institutions. The results of this research indicate that the samples can be processed faster, while allowing for decreased reagent consumption and, consequently, at a reduced cost. The concordance profiles obtained with different amplification kits indicate compatibility between various primers and amplification reagents.

Improving Throughput, DNA Typing, Swabs/Reagents

A80 Enhancing the Sexual Assault Workflow: Testing of Next Generation DNA Assessment and Y-STR Systems

John Ballantyne, PhD; Erin K. Hanson, PhD; Robert L. Green, BA; Allison Holt, PhD; Julio J. Mulero, PhD*

After attending this presentation, attendees will learn how the newly developed next generation DNA quantification and Y-chromosome Short Tandem Repeat (Y-STR) kits can improve the analysis of challenging sexual assault evidence.

This presentation will impact the forensic science community by presenting possible improvements to the sexual assault workflow processes in operational forensic science crime laboratories.

Sexual assault samples are among the most difficult sample types encountered by forensic laboratories. Typically, a sexual assault sample has multiple challenges including small quantity of male DNA, relatively high quantity of female DNA, and presence of Polymerase Chain Reaction (PCR) inhibitors. These factors make it difficult to obtain an interpretable male profile. Therefore, there is a need for more robust, highly sensitive, and faster methods for the assessment (i.e., quality and quantity) of DNA extracts to determine optimal downstream processing methods, as well as improved Y-STR amplification systems for profiling these difficult samples.

Studies were conducted to evaluate the efficacy of a new Y-STR kit (Yfiler® Plus) and a new DNA quantification kit (Quantifiler® Trio) in addressing these needs compared to

previously used methods. This next generation Y-STR kit includes 27 markers (including 7 rapidly mutating Y-STR loci) with optimized amplification balance. Full profiles are attainable with high levels of commonly encountered PCR inhibitors (e.g., hematin and humic acid) and low levels of male DNA in the presence of high quantities of female DNA (e.g., 1:1000 and 1:4000). The workflow improvements reduced amplification run time to less than 1.5 hours.

The new DNA quantification kit is designed to generate more accurate information about casework samples, and from sexual assault samples in particular. This next generation DNA quantification kit exhibits high sensitivity, in the picogram level (~2 pg), for both the human and male targets, higher inhibitor tolerance to match the next generation Y-STR kit, and additional critical tools (e.g., male/female ratio calculation and a degradation index) for the determination of DNA quality. The time required to set up the assay and perform amplification has been reduced to less than one hour. The current studies presented here indicated that the new DNA quantification and sample assessment kit provides better correlation with more recently released STR kits like the new Yfiler® Plus kit.

These two next generation systems can generate an improved workflow for obtaining interpretable profiles from sexual assault samples. The utility of the new workflow, as compared to previously used methods, in processing challenging sexual assault samples will be presented. The successful use of the next generation DNA quantification and assessment kit, as well as the Yfiler® Plus Kit, to obtain informative Y-STR profiles from challenging sexual assault sample types, specifically samples collected four, seven, and nine days after intercourse, will be demonstrated. Despite the presence of low quantities of male DNA and female DNA in vast excess (1:333 male:female ratios or greater), probative Y-STR profiles were obtained from these samples using the next generation quantification and amplification systems with mostly full profiles obtained if >100pg of male DNA was present. Additionally, a good correlation between male quantitation and profile recovery was observed, with a negative quantitation value indicating that a negative or unusable profile would be obtained.

Y-STR Analysis, Sexual Assault Evidence, Next Generation Y-STR Kits

A81 How Low Can You Go? An Evaluation of Reduced Reaction Volumes for Direct Amplification With PowerPlex® Fusion

Daniel Watsula, MS; Jon Davoren, MS; Jangbir Sangha, MA*

After attending this presentation, attendees will gain an understanding of how to achieve a high first pass success rate when performing direct amplification of 1.2mm punches from buccal samples collected on nontreated paper with PowerPlex® Fusion. Attendees will also learn that a high first pass success rate can be achieved with reduced reaction volumes in addition to the standard manufacturer's recommendations.

This presentation will impact the forensic science community by demonstrating the feasibility of reduced volume direct amplification with PowerPlex® Fusion for the processing of reference samples.

The efficiency of reference sample processing for databasing and paternity purposes has been greatly increased by the development of direct amplification systems. DNA samples can be collected and stored on non-treated matrices, such as the Bode Buccal DNA Collector™, until sample processing is required. The Promega® Corporation has recently released a new direct amplification kit encompassing 24 loci. PowerPlex® Fusion was designed for direct amplification of samples collected on both

treated and non-treated collection paper. Direct amplification of reference samples eliminates the extraction and quantification steps encountered during routine DNA analysis as an effort to save time and costs in the laboratory.

This presentation will describe the studies with PowerPlex® Fusion to obtain optimal results from samples collected utilizing the Bode Buccal DNA Collector™, a non-treated matrix, at varying reaction volumes. The amplification reaction volumes analyzed in this experiment were 25µL (full reaction), 12.5µL (half reaction), 6µL (quarter reaction), and 15µL. A total of one hundred (n=100) self-collected samples obtained in either 2010 or 2011 (2- to 3-years-old at time of testing) were utilized in this experiment. Collectors from 2010 and 2011 were chosen as they may be more indicative of a sample encountered during routine databasing rather than a fresh sample collected a few days prior to testing.

This presentation will display the optimized procedures for cell lysis, reaction mix components, thermal cycling parameters, and 3130XL injection conditions. Amplification reaction mix components were optimized to overcome potential inhibitory effects of the lysis solution. When the lysis solution is not completely evaporated and inactivated, it can result in faint or absent allele peaks.¹ As the reaction volume decreased, the relative concentration of the lysis solution in the amplification reaction increased, which may also cause inhibition. To overcome this potential inhibition at the reduced volume reactions, 5X AmpSolution™ Reagent was added to the reaction mix. These optimized procedures resulted in a first pass success rate of 84% for a full reaction (25µL). The highest first pass success rate (95%) was observed when utilizing the half reaction (12.5µL) protocol. The quarter reaction protocol (6µL) also resulted in a first pass success rate of 84%, while the 15µL reaction resulted in an 89% first pass success rate. In addition to displaying first pass success rates for each reaction volume, interlocus balance information for each will also be discussed. Results of research evaluating the viability of a 3µL PowerPlex® Fusion reaction with a sample from a non-treated collection paper sample matrix will also be presented and discussed.

While eliminating the extraction and quantification steps with direct amplification can save time, if normalization does not occur, it can also lead to additional amplification reactions and costs. Cellular deposition can vary between individuals resulting in either excessive or inadequate samples. Eating, drinking, medicine intake, health status, and non-cooperation during sample collection will all affect the amount of cells deposited onto the collection paper. This variation in cellular deposition can either lead to over- or under-amplification if the procedure is not optimized. Optimizing the amplification parameters to account for sample variation reduces re-sampling and re-amplification. This reduction in reprocessing can result in an improvement in the overall processing efficiency of the laboratory.

Direct amplification of reference samples can provide a time efficient method for obtaining complete genetic profiles but can be cost prohibitive. Utilizing one of the reduced volume reaction procedures demonstrated in this presentation can greatly reduce the cost per amplification reaction. This may allow for the process to not only be time efficient but also cost efficient.

Reference:

1. Promega PunchSolution™ Kit Technical Manual: <http://www.promega.com/~media/files/resources/protocols/technical%20manuals/101/punchsolution%20kit%20protocol.pdf?la=en>

Direct Amplification, PowerPlex® Fusion, Reduced Volume Reactions

A82 Predicting the Quality of DNA Profiles Through the Evaluation of the ParaDNA®

Screening Instrument

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After attending this presentation, attendees will have an understanding of the ParaDNA® Screening Instrument and its applications in the forensic DNA laboratory as a screening tool to predict the quality of DNA profiles.

This presentation will impact the forensic science community by providing a correlation between results given by the ParaDNA® Screening Instrument and traditional Short Tandem Repeat (STR) analysis, potentially allowing a forensic laboratory to select samples that may generate higher quality DNA profiles while minimizing the analysis of samples with low quality or quantities of DNA.

Forensic DNA laboratories throughout the United States are experiencing a backlog of casework due to an influx of requests to process all cases that contain biological evidence. DNA analysis is a time-consuming and expensive process and in some instances a sample may not yield an interpretable profile or even a quantifiable amount of DNA. In these cases, it may be necessary to analyze a large number of samples in order to obtain a useable DNA profile for comparison. In order to determine if the laboratory could limit the number of low-quality samples it processed, the Palm Beach County Sheriff's Office (PBSO) Forensic Biology Unit evaluated the LGC Forensics ParaDNA® Screening Instrument with the Screening Kit. The ParaDNA® instrument processes evidentiary samples in 75 minutes, reporting a percent score indicative of whether the sample will yield a positive result from further laboratory profiling and whether the DNA originates from a male or female.

In order to determine if the ParaDNA® instrument could be implemented at PBSO to screen and reduce the number of samples analyzed for DNA, 82 samples were analyzed on the ParaDNA® instrument and processed through current PBSO protocols. These mock evidentiary samples represented the major types of samples encountered in casework: blood; saliva; touch; and, mixed semen samples. After analysis, samples were categorized based on their "percent score" as determined by the ParaDNA® software as follows: 75-100%; 25-74%; 1-24%; and, 0%. Conventional sample analysis was then conducted using the Qiagen® EZ1® Advanced XL and the DNA Investigator® Kit for extraction, Promega® Plexor® HY and an Applied Biosystems® 7500 Real-Time PCR System for quantification, Promega® PowerPlex® 16 and an Applied Biosystems® GeneAmp® PCR System 9700 for amplification, an Applied Biosystems® 3130xl Genetic Analyzer, and Applied Biosystems® GeneMapper® ID-X Software for analysis. The ParaDNA® percent score of each sample was compared to the quantification value of the sample, the quality of the DNA profile obtained (full, partial, full mixture, partial mixture, or negative) and the average Relative Fluorescence Units (RFU) value for each dye channel to determine if the profile was above or below stochastic threshold. These comparisons were used to determine the correlation between the ParaDNA® percent score and the expected STR profile result.

The results of the evaluation showed that for all samples the ParaDNA® instrument rated greater than 75%, the quantification value was greater than the target amplification value of 0.8ng, and resulted in a full or full mixture profile. Of the samples rated between 25% and 74%, only 33% were above the target amplification value,

yet 91% of these samples produced full or full mixture profiles. Only 4% of samples rated between 1% and 24% were above the target amplification value, with 46% resulting in full profiles. None of the samples rated 0% were above the target amplification value, yet 36% were able to yield a full profile; however, the majority of these profiles were below the laboratory's stochastic threshold. Future studies at PBSO should include additional samples to further enhance the correlations found by this study, as well as evaluate the Intelligence® Kit. The results of this evaluation indicated that the ParaDNA® Screening Instrument with Screening Kit may serve as a useful tool to prioritize evidentiary samples to be processed for STR analysis by helping to determine which samples may yield the most interpretable DNA profiles.

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DNA Screening, ParaDNA, DNA Interpretation

A83 A Proposed Training Program for DNA Technical Leads Using Learner-Centered Instruction

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The goal of this presentation is to provide a better understanding of how to design courses that emphasize the application of learned material to real-life situations.

This presentation will impact the forensic science community by making those responsible for forensic science instruction aware of basic concepts of instructional design and systematic ways to develop course material.

Traditional methods of instruction have presented information as separate modules of learning with sometimes little integration of course content from one module to the next. Instruction may be disjointed and the presentation of isolated facts does not always promote long-term recall of the material. When instructional content is linked to real-world applications, the content of the class takes on new meaning, allowing students to take charge of their knowledge and apply it in ways that will be useful to them in their future careers.

The proposed training program will cover a number of topics that are of relevance to the duties of a DNA Technical Lead including: (1) principles of creative leadership, such as fostering innovation, running productive meetings, strategic planning, and providing efficiency in DNA operation (case assignment, Sigma Six, and flow analysis); (2) managing projects; (3) DNA technical leadership which incorporates improving DNA interpretation through writing guidelines and procedures; (4) job aid construction; (5) validation of methods and instruments such as DNA automation (robotics); (6) grant management comprised of budgeting, time management, and writing grant proposals; (7) training/instructional design including mentoring; (8) quality control/quality assurance, encompassing DNA/ASCLD inspections and root cause analysis; (9) innovative approaches to casework such as reviewing old cases with new technology; (10) report reviews; and, (11) Combined DNA Index System (CODIS) Administration.

Learner-centered instruction is designed with the end goal in mind based on formulation of key concepts. Important factual knowledge (e.g., DNA interpretation principles, CODIS regulations, etc.) and discrete skills (e.g., use of statistics, spreadsheet use, error analysis methodologies, etc.) are taught in a sequence to make

them applicable to each lesson content. Transferable skills that can be related from one major topic to the next (e.g., use and evaluation of research data bases, mentoring/instructional planning, etc.), and more complex processes (e.g., creative leadership, innovative approaches to case work, etc.) are integrated into the lesson plans which are developed to supplement lecture and provide a means of practicing the techniques and applying them to new situations. From this synthesis of ideas, one can develop a few central themes (e.g., leadership and innovation, technical competence, validation, task analysis, instruction and mentorship) by which the individual modules of instruction can be linked.

Designing learner-centered instructional material is difficult.¹ Even for experienced professionals, sorting out the key aspects of laboratory operation, presenting them in a logical fashion, and designing instructional content around these central ideas is difficult. Taking advantage of published worksheets and training aids for developing material for classroom presentation can be helpful. The technical facts and the sometimes abstract central concepts can then take on new meaning when they can be integrated into some type of real-life performance strategy.^{2,3}

Assessment of student learning is an important aspect of course design which has traditionally been measured by multiple-choice questions which have emphasized lower level skills, such as retention of isolated facts and concepts, at the expense of higher-order skills, such as reasoning and evaluation. Once key skills are identified, they can be used to identify key underlying concepts, and the relative value of these concepts can be assessed to determine the cognitive level at which test questions should be asked, strategies for improving performance can be implemented, and contexts in which the skills are to be utilized can be identified.

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3. Wiggins, Grant and Jay McTighe (2004), *Understanding by Design Professional Development Workbook*. Alexandria, VA: Association for Supervision and Curriculum Development.

DNA, Instruction, Validation

A84 Trends of Small DNA Units in Local Jurisdictions: Pros and Cons

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After attending this presentation, attendees will be informed of both the obvious and the often-overlooked issues facing small DNA units within local jurisdiction laboratories. Data will be shared regarding trends in the number of DNA requests, cost, inherent advantages and disadvantages, staffing requirements, and how common challenges are amplified by limited staffing.

This presentation will impact the forensic science community by providing valuable metrics to those jurisdictions considering adding DNA units to existing laboratories. The Colorado Springs Police Department Metropolitan Forensic Laboratory DNA Unit has been in operation since the fall of 2008. This retrospective study is a historical and candidly informative view of the first five years post-implementation.

The increased demand for rapid-turnaround DNA testing by state forensic laboratories has led to a greater backlog of cases and much slower turnaround times. Many local jurisdictions are

now adding DNA units to their existing local crime laboratories in the attempt to reduce turnaround times, backlogs, and to gain investigative information while cases are still warm. Due to the infrequency of new DNA units being created, often local jurisdictions have limited resources available to them for planning and to accurately forecast various needs. Attendees of this presentation will be provided a five-year retrospective of the design and execution of a two-person DNA unit to support investigations for a population of approximately 640,000 people in a land mass of greater than 2,100 square miles. A detailed assessment of casework produced between 2008 and 2013 will demonstrate the unit's overall administrative and quality-assurance functionality, and will determine if the long-term goals have been met.

In 2001, the first serious discussions occurred regarding the creation and implementation of a DNA unit within the Colorado Springs Police Department Crime Laboratory. Initial funding for the unit was provided in 2004 and 2005 from the department's general fund. The startup of the Colorado Springs Police Department Crime Laboratory would not have been possible without the fund-matching grants funded by National Institute of Justice (NIJ) Award #2004-RC-CX-K019 awarded by the National Institute of Justice, Office of Justice Programs, U.S. Department of Justice. Additional funding was appropriated from local Public Safety Sales Tax (PSST) in 2006, which was the same year renovation began of existing laboratory space and the DNA technical leader was hired. Renovation was completed at the end of 2007 and a second DNA analyst was hired. Validation of chemistries and instrumentation were performed and the unit was accredited by Forensic Quality Services (FQS) to the International Organization for Standardization/International Electrotechnical Commission (ISO/IEC) 17025:2005 standards in October 2008. Forensic DNA laboratories are required to have at least two DNA analysts, one of which must serve as the DNA Technical Leader. Staffing shortfalls in the DNA unit occurred in 2010, 2011, and 2012, with a resulting dramatic reduction in cases completed during that time span.

This presentation will provide much-needed metrics to illustrate the administrative burden of validating and implementing new technologies, maintenance of quality assurance and accreditation requirements, and the realistic capabilities of casework completion for a minimally-staffed, two-person serology and DNA unit. In addition, a historical evaluation of planning, cost analysis, and investment of time and resources will be discussed. A discussion of the benefits and challenges of grant funding will be included (free money is never free — there is always a cost). The presentation will also touch on what happens when you're the victim of your own success. Finally, the inherent benefits of working closely with investigators will be described to benefit other small jurisdictions.

The opinions, findings, and conclusions or recommendations expressed in this presentation are strictly those of the author and do not necessarily reflect those of the National Institute of Justice or the Colorado Springs Police Department Metro Crime Laboratory.

Small DNA lab, Pros and Cons, Local Jurisdiction

A85 Real-Time DNA Analysis at the Crime Scene From the Perspective of the Crime Scene Officer

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After attending this presentation, attendees will have insight concerning how the future introduction of mobile DNA technologies will change the work of the crime scene officer and what impact this technology can have on the criminal justice system.

This presentation will impact the forensic science community by providing information on the current and future process for the search, collection, and selection processes of DNA traces and how analysis results from real-time mobile technologies are handled by the crime scene officer.

It is anticipated that mobile technologies for real-time DNA analysis on the crime scene will become available in the near future. The decision-making process on the actual introduction of mobile DNA technologies in criminal investigations is, among others, dependent on the performance characteristics of the existing DNA analysis technology. A crucial performance characteristic of any forensic analysis system is the turnaround time. By evaluating the criminal files from 67 serious and 176 high-volume crime cases in one of the police regions in the Netherlands, the actual DNA analysis turnaround time was assessed. The results of this evaluation show an average DNA analysis turnaround time of 66 days for traces from the serious crime trajectory and 44 days for traces from high volume crime trajectory. The data also show that a relatively large number of crime scene samples that have been secured and submitted for DNA analysis fail to produce informative DNA-profiling data. Thirty-eight percent of the samples from the serious crime trajectory and 17% of the samples from high-volume crime trajectory generated no DNA-typing results.

Before the actual implementation of mobile DNA technologies, it is important to understand how this technology can be optimally integrated in the work of the crime scene officer. To be able to compare the processes at current and future crime scene investigations, observation studies were performed in a mock crime scene. Crime scene officers in the experimental group (n=20) had the opportunity to make use of fast DNA-analysis, whereas the crime scene officers in the control group (n=20) performed their research under normal conditions. The results show that there is a positive attitude of the crime scene officer toward mobile DNA technologies. There were no significant differences between the number of traces that were secured by the two groups from the mock crime scene; however, the number of samples that was analyzed through real-time DNA analysis by the experimental group exceeded the number of samples that were selected by the control group for laboratory analysis.

The results of this study are presented in the context of the main goal of this study: to create a comprehensive platform that allows for the introduction of mobile DNA technologies.

Real-Time DNA Analysis, Crime Scene Officer, Forensic Process

A86 Back to the Start: Visual Attention of Crime Scene Officers During Their Exploratory Rounds and the Influence of Mobile Identification Techniques on the Perception of Traces

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After attending this presentation, attendees will learn about visual attention processes of crime scene officers during their explanatory rounds and the influence of fast identification techniques that can be used at the crime scene on these processes.

This presentation will impact the forensic science community by contributing to the understanding of basic visual attention processes at the crime scene. Therefore, it provides a base of evidence for developing the methods of crime scene investigation and for making informed decisions about implementing new rapid identification techniques. The ultimate goal is to improve the quality of crime scene investigations. Crime scene investigation plays a crucial role in police investigations, yet the body of evidence in forensic science is primarily focused on technology and laboratory processes that follow the crime scene investigation and less on the crime scene investigation itself.

In the near future, technological developments will make it possible to conduct DNA, chemical and fingerprint analyses at the crime scene, and to directly compare obtained profiles to a database. This development will certainly speed up the investigation, but it will also entail risks, because the investigation might more easily focus on the wrong subject when the analyzed traces are not related to the crime. A focus on certain traces, and consequently on a certain person, might be already developed during the first phase of the investigation. The interpretation of the scene and its traces already starts during the first, exploratory round in which the crime scene officers observe the scene and after which they draw up a plan based on what they have seen. The availability of, for example, a fast DNA technique might change the focus of the officer, with the consequence of looking for biological traces to conduct fast DNA analysis without reconstructing what these traces mean and without considering their relevance. The same goes for fingerprint techniques or chemical techniques such as a spectral camera: the focus might become focused more on fingerprints at the crime scene or possible blood traces and not on the initial reconstruction of the (alleged) criminal act through verification and falsification. So, the effect of the new technologies might be that attention of the crime scene officers changes, depending on the technology available. In order to understand visual attention and the influence of fast identification techniques, a study was conducted in a mock crime scene with 30 crime scene officers. Three conditions were created: (1) the control group conducted a traditional investigation; (2) the second group was able to use fast identification techniques for biological traces and fingerprints; and, (3) the third group was able to use a device, a spectral camera, for fast chemical analysis of traces and could get an indication of the age (time since deposition) of blood stains. During their investigations, the crime scene officers were observed. In order to gain insight into their thought processes and attention processes, officers worked with a trainee (one of the researchers). The data consist of video and audio recordings of the crime scene investigations, information about thought processes and attention processes of the investigators, and interviews conducted after the investigation was finished. The results show how traces are perceived and judged by the different groups of crime scene officers.

Crime Scene Investigation, Fast Identification Techniques, Visual Attention

A87 Latent Heat Thermography Imaging: A New Visualization Tool for Forensic Science

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The goal of this presentation is to present studies establishing the scientific basis and practical application of latent heat thermography for detection of bloodstains in the presence of other background materials that might be present at a crime scene. Attendees will learn about the development of a novel and sensitive infrared system for rapid and nondestructive visualization of bloodstains.

This presentation will impact the forensic science community by describing a simple method for nondestructively imaging dilute (1,000X) latent bloodstains on cloth with a thermal infrared camera following exposure of the fabric to water vapor.

An important task in a criminal forensic examination is to identify potential biological fluid stains on materials of evidentiary value (e.g., clothing from a crime scene). The most sensitive available methods (e.g., luminol) risk damaging and contaminating DNA evidence, while the most nondestructive (e.g., alternate light sources) are also among the least sensitive. Infrared imaging modalities are being developed to visualize blood and other bodily fluids on fabrics and other substrates. Developing techniques for the visualization of biological fluid stains on common surfaces has been a continuing goal for numerous forensic studies.¹⁻³ The ideal device would be small, relatively inexpensive, easy to operate and maintain, and portable; enhancement reagents would not be necessary; the method would be nondestructive; further, the device would detect trace levels of blood and operate indoors or outdoors under ambient lighting. It is well known that fabrics adsorb/desorb water depending on humidity. Differential contrast was observed in the thermal response of bloodstained and clean fabrics on exposure to water vapor generated from a hand-held steamer of the type used by travelers to remove wrinkles from garments. Because this phenomenon is related to the enthalpy of adsorption of water vapor, the technique is called Latent Heat Thermography (LHT) imaging. Evidence suggests that the mechanism of visualization is the differential rate and extent of vapor absorption. The present scope of this study has been to find conditions that best visualize bloodstains on fabrics, and to understand the mechanism(s) by which dilute bloodstained fabric is distinguished from unstained fabric.

Initial testing used two fabric samples. The first was a coarse, dyed, unpatterned acrylic fabric with symbols drawn using diluted rat blood. The symbol "I" in undiluted blood was easily observed; "X" in 10-fold diluted blood and "V" in a 25-fold diluted blood were also observable; the "L" and a "C" drawn in 50- and 100-fold dilutions of blood on the second line were not visually observable. The second fabric sample tested was black polyester with hexagonal glittery skull-and-crossbones appliques clearly visible from both sides. This fabric has two handprints created by a latex-gloved right hand pressed into a shallow pan of blood solution and then laid on the fabric. Neither handprint was visually observable. One handprint was made with blood diluted by a factor of 100 with water, the other with blood diluted by a factor of 1,000

with water.

A trained forensic scientist with the South Carolina State Law Enforcement Division using two alternate light sources for blood detection was able to observe stains down to 100X dilution on the unpatterned acrylic fabric. Neither of the stains on the patterned polyester fabric were detected by the investigator, even knowing that they were present and observing a repetition of the vapor exposure experiment in person. All stains were easily visualized by steaming with water vapor, illuminating the target with Infrared (IR) light, and observing the light reflected from the samples. Data processing included lock-in amplification and image processing to enhance discrimination as done in earlier work. Other experiments have shown that stains of materials having varying hydrophobicity show contrast, and it is expected that vapors with chemical properties distinct from water would show different effects from those observed with water. Likewise, there is no reason to believe that fabrics are the only substrates on which stains or coatings are observable, but this has not been investigated to date.

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Blood Detection, Thermal Imaging, Bloodstain Visualization

A88 Identification of Body Fluid Traces Using Raman Microspectroscopy: Substrate Interference and Contaminations

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The goal of this presentation is to describe the development of a novel method for nondestructive, confirmatory identification of body fluid traces on the crime scene and in the laboratory. Specific attention was focused on the most recent development of the method for characterization of contaminated samples and substrate interference. After attending this presentation, attendees will have a better understanding of the recent advances of this application of Raman spectroscopy. The implementation of advanced statistics for automated analysis of spectroscopic data and the evaluation of the accuracy and reliability of the conclusions made will be discussed.

This presentation will impact the forensic science community by improving the accuracy and effectiveness of biological stain analysis for forensic purposes.

The identification of traces of body fluids discovered at a crime scene is a major part of forensic investigation today. The three most common fluids found are blood, semen, and saliva, and there are several methods used currently to distinguish one from another. Blood can be presumptively tested by using different color spot tests, but these tests are destructive to the sample and can also have false positives. Semen is similar in that there are destructive presumptive tests as well as confirmatory tests. Saliva,

however, has no confirmatory tests. Most presumptive tests can be performed in the field, but some sample preparation such as extraction is often necessary. Most confirmatory tests must be done in the laboratory. The main problem with these tests is the destruction of the sample. The forensic science community is in need of a reliable, non-destructive, field method for identification of all common body fluids.

Raman spectroscopy is a technique that is increasing in popularity among the different disciplines of forensic science. Some examples of its use today involve the identification of drugs, lipsticks, and fibers, as well as paint and ink analysis. The theory behind Raman spectroscopy is based on the inelastic scattering of low-intensity, nondestructive laser light by a solid, liquid, or gas sample. Very little or no sample preparation is needed, and the required amount of material tested with a Raman microscope can be as low as several picograms or femtoliters. A typical Raman spectrum consists of several narrow bands and provides a unique vibrational signature of the material. Typically, nonresonance Raman spectroscopic measurements do not damage the sample. The stain could be tested in the field and still be available for further use in the laboratory for DNA analysis. A portable Raman spectrometer is a reality now that should allow the identification at the crime scene.

This study concerns the latest development of a new method for identification of body fluid traces using Raman spectroscopy combined with advanced statistics. Multidimensional Raman spectroscopic signatures of dry traces of sweat, vaginal fluid, semen, saliva, and blood have been reported earlier. The ability of the method to detect and identify small amounts of semen and blood in mixed samples has been also demonstrated. A method that allows for differentiating menstrual and peripheral blood will be presented.¹ Combined software for the identification of all major body fluids and the evaluation of the accuracy and reliability of the conclusions made will be demonstrated. The method was expanded for the application to blood and semen samples contaminated heavily with sand, dust, and soil.² Potential interference of commonly encountered substrates will be discussed.³

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Body Fluid, Raman Spectroscopy, Statistical Analysis

A89 Rapid, On-Site, Body Fluid Identification and Detection by Surface Enhanced Raman Spectroscopy (SERS) for Forensic Applications

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After attending this presentation, attendees will be able to assess and appreciate the significant advantages that the techniques of Surface Enhanced Raman Spectroscopy (SERS) will bring to crime scene investigations for the rapid, on-site identification of human body fluids such as blood, semen, urine, saliva, vaginal fluid, and body fluid mixtures.

This presentation will impact the forensic science community by introducing a well-established analytical spectroscopic technique for the rapid, on-site, sensitive confirmatory identification of human body fluids. The capability of this optical methodology has never previously been demonstrated, and offers several advantages relative to current confirmatory methodologies in terms of ease-of-use, cost, sensitivity, and specificity.

Body fluids are among the most important and most common evidence collected at crime scenes. New and developing technologies can provide forensic crime investigators with increasingly powerful methodologies for more accurate and increasingly rapid means for identifying body fluids. In turn, such advances contribute to faster confirmation of crime scene events, crime time lines and perpetrator apprehension. This presentation will illustrate the fundamental capabilities of SERS for the detection, identification, and characterization of trace amounts of blood, semen, vaginal fluid, saliva, and their mixtures for forensic purposes. In contrast to current forensic science techniques, subsequent development of this methodology can lead to a single optical platform for the rapid, sensitive, easy-to-use, cost-effective, on-site, non-destructive detection and identification of human body fluids at a crime scene. No such platform is currently available for this purpose. Furthermore, although Raman spectroscopy has just recently been employed to address the challenges of body fluid identification for forensic science applications, SERS has not been previously used as a forensic tool for trace body fluid analysis and offers significant advantages, as will be demonstrated here.¹⁻⁷

Raman scattering, the inelastic scattering of incident laser light resulting in unique vibrational signatures due to the molecular components in sample mixtures, is a well-known optical technique that is ideally suited for many bioanalytical applications owing to its high-information content, speed, ease-of-use, portability, and label-free character; however, it is a relatively weak effect. The use of SERS overcomes this limitation by enhancing Raman efficiencies by typically 10^6 - 10^8 for molecules close to nanostructured metal surfaces, thus making this Raman probe exquisitely sensitive to some analytes at low concentration as well as enhancing molecular specificity.⁸ Both of these attributes play a large role in this application for the detection and identification of trace amounts of body fluids. Further advantages of SERS relative to normal Raman for forensic science include no fluorescence and greater signal reproducibility.

High-quality SERS spectra of dried human blood, semen, vaginal fluid, saliva, and urine have been obtained and will be displayed. All of these body fluids exhibit unique SERS vibrational signatures. These spectra are acquired on previously developed, *in-situ* grown, Au nanoparticle-covered SiO₂ substrates and identification is accomplished by the previously developed second derivative based "barcode" statistical analysis methodology.^{9,10} These SERS substrates are inexpensive to produce, provide strong signal enhancement, and have been successfully used for a number of bioanalytical identification applications. Blood has been extensively studied by Raman for forensic applications, but

improvements in sensitivity and homogeneity (spatial and spectral) resulting from SERS will be shown and lead to greatly improved Raman-based blood identification capabilities.^{1,3,6,7} In addition to trace blood detection and identification, SERS may also be used for bloodstain age determinations. Distinctions between human, dog, rat, and cow blood will be described as well.

Preliminary results show that SERS offers more consistent results for saliva, semen, and vaginal fluid identification than normal Raman, provides a distinction between spermic and aspermic samples, and can identify mixtures of vaginal fluid and sperm.^{2,4,5} SERS data acquisition is accomplished in the time frame of minutes, and the use of a portable Raman device such as the unit built for this research allows these procedures to be carried out at crime scenes. Plans for the subsequent development of this methodology will be described.

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SERS, Body Fluids, Confirmatory Identification

A90 Toward a Portable, Universal Mass Spectrometer: Simultaneous Elemental, Isotopic, Inorganic, and Organic Analyses of Forensic Samples

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After attending this presentation, attendees will gain an

understanding about current research efforts toward development of field-portable mass spectrometers capable of “universal” analysis and an appreciation for the capabilities of ambient mass spectrometry.

This presentation will impact the forensic science community by providing a method for simultaneously acquiring isotopic, elemental, and molecular information about a sample using ambient ionization mass spectrometry. This method furthers efforts in bringing mass spectrometers directly to crime scenes to provide comprehensive, real-time analysis.

When the chemical composition of evidence is studied, it can be analyzed on different instruments dedicated to determining a specific aspect of the sample. If elemental or isotopic information is needed, typically Inductively Coupled Plasma/Mass Spectrometry (ICP/MS), X-Ray Fluorescence (XRF), or Scanning Electron Microscopy/Energy-Dispersive X-Ray Spectroscopy (SEM/EDX) are used and to acquire molecular information about the sample, it is analyzed separately using a method such as Liquid Chromatography/Mass Spectrometry (LC/MS), Gas Chromatography/Mass Spectrometry (GC/MS), or Fourier Transform Infrared Spectroscopy (FTIR). Recent research has shown that combining elemental (e.g., lead, barium, antimony) and molecular information (e.g., diphenylamine) about gunshot residue enables enhanced GSR information; analysts using this technique could differentiate the caliber of weapon from which the GSR originated.¹ Simultaneous analysis of multiple aspects of a sample with a mass spectrometer could be a powerful tool in forensics, combining aspects of trace analysis (elemental composition) with chemistry (molecular properties). To facilitate such analyses with mass spectrometry, appropriate ionization sources need to be developed and tested.

In these experiments, Desorption Electrospray Ionization (DESI) and Direct Analysis In Real Time (DART®) were assessed as soft ambient ionization techniques for acquiring mass spectra of molecular and soluble inorganic species while laser ablation was studied for determination of elemental information. The ionization sources were coupled to an ion trap mass spectrometer. While all of these experiments were performed in a laboratory setting, the methods were chosen such that they were amenable to portable instrumentation; DESI and DART® have previously been used with portable ion trap mass spectrometers for field work.^{2,3}

This research focused on samples relevant to gunshot residue, explosives, and nuclear forensics. Using DESI and DART®, common organic and inorganic explosive compounds were analyzed, while laser ablation was used to acquire elemental and isotopic information. Both organic gunshot residues and metal primer components were detected using this method. The error in isotopic ratio measurements was ~10%, which is not accurate enough for determining variation in stable isotopes, but is adequate for nuclear forensics to determine levels of isotopic enrichment for radionuclides. In testing mock Radiological Dispersion Devices (“dirty bombs,” RDDs), both the explosives used for dispersion (molecular) and the radionuclides (elemental/isotopic) were detected in one analysis.

In conclusion, by combining soft (DART® and DESI) and harsh (laser ablation) ionization techniques, elemental, isotopic, and molecular information was acquired simultaneously. Further, the methods used are readily amenable to incorporation in portable instrumentation.

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Ambient Mass Spectrometry, Laser Ablation, Nuclear Forensics

A91 *In Situ* Differentiation and Identification of Perylene Pigments in Automotive Finishes Using Extended Range (4000-250cm⁻¹) Fourier Transform Infrared (FTIR) Spectroscopy

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The goal of this presentation is to enable the identification of perylene pigments *in situ* in automotive and other types of paint using infrared spectroscopy. This can be useful for classification, differentiation, and identification of automotive paint.

This presentation will impact the forensic science community by providing forensic paint examiners with information regarding identification of some pigments used in red and brown metallic and nonmetallic automotive finishes, as well as other materials that might be encountered as trace evidence.

The perylenes are a family of high-performance organic pigments that occur mostly in red, violet, and brown shades. Owing to their high color strengths, good thermal stabilities, and excellent lightfastness and weather-fastness properties, they are very common in automotive finishes. Perylenes were commercially introduced in the 1950s and Perylene Maroon (Pigment Red 179) and Perylene Red (Pigment Red 178) were first marketed in 1959 and 1966, respectively.

Seven perylenes have been cited as automotive paint pigments by various authors, but only five of these, Perylene Maroon (Pigment Red 179), Perylene Red (Pigment Red 178), Perylene Vermillion (Pigment Red 123), Perylene Red Y (Pigment Red 224), and Perylene Bordeaux (Pigment Violet 29), were being marketed for this application when this study began in 1997. The purpose of this investigation was to determine which of the five are actually used in automobile Original Equipment Manufacturer (OEM) finishes (1974 to present), and how they can be differentiated and identified *in situ* based on their infrared absorptions in paint spectra (4,000 to 250cm⁻¹). Usage information, including the colors and the types of paints (metallic or nonmetallic) they are found in, the commonalities of their use, whether they occur as masstones (that is, undiluted without other pigments present), which other pigments they are typically combined with, and if relevant, approximate time frames in which they were used, was then sought.

Perylene Red Y appears to be unique among organic pigments in having an acid anhydride functional group, and this moiety is responsible for the strongest absorption of this pigment, which occurs at 1,774cm⁻¹. This is higher in frequency than the binder ester carbonyl absorption (1,735cm⁻¹), and because it is so unusual and conspicuous, this one absorption can often serve — by itself — to indicate the presence of this pigment. Perylene Red Y was much more prevalent in monocoats than in basecoats and use of this pigment has been declining because it is prone to hydrolysis in alkaline solutions. Consequently, it is not formulated into water-based basecoats, which are becoming more widely used

due to environmental regulations. Perylene Red Y is used in both metallic and nonmetallic finishes, although it is more common in the former.

Perylene Maroon was found to be, by far, the most common perylene, and it was identified in 87 of the 98 (89%) red or maroon metallic basecoats (1977 to present) examined in this study. It is used predominately in metallic finishes, although it was also identified in a few nonmetallic red or maroon basecoats.

Perylene Red was only identified in a dozen or so nonmetallic monocoats and basecoats, mostly from the late 1980s. It is no longer used much and was largely supplanted by another organic pigment, DPP Red BO. Perylene Bordeaux is used in both metallic and nonmetallic finishes and was found in a dozen dark maroon finishes. Perylene Vermillion was not identified in any finishes examined in this study.

A large number of different pigment combinations involving perylenes were identified in this study. They include formulations of perylenes combined with each other (Perylene Maroon with either Perylene Red Y or Perylene Bordeaux) and with other color-imparting organic (quinacridones), inorganic (ferric oxide), and effect pigments (Red Mica), as well as with inorganic extender pigments (barium sulfate and kaolin). These different compositions result in infrared spectra that exhibit a very wide diversity of absorption patterns and examples of this diversity are presented and discussed.

Paint, FTIR, Perylenes

A92 The Utilization of Forensic Science Principles and Microscopic Trace Evidence Techniques to Unequivocally Prove That a Long-Suspected Painting by Jackson Pollock Was Crafted at the Pollock-Krasner Home and Studio — a National Historic Landmark

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After attending this presentation, attendees will: (1) know how to use trace evidence methods to authenticate works of fine art; and, (2) learn a new approach to the unbiased verification of questioned works of art.

This presentation will impact the forensic science community by presenting new genera for the use of forensic trace evidence analytical techniques in the certification of works of fine art.

Normally, questioned works of art are authenticated by considering an array of factors. First, an object's or painting's history of ownership or provenance is typically of prominent importance. Next, features such as an artist's technique, marks, patterns, and designs formed by placement of the medium with a palette knife or paint brush are crucial to their endorsement. Other important aspects including microscopic and chemical analyses of the types of materials (i.e., canvas, paint pigments, and resins)

used to produce a work are vital to its legitimacy. Fractal analysis has also been employed to authenticate some artists' works. Often, less scientific forms of proof, such as the analysis of an artist's signature or the identification of a partial fingerprint, leads only to confusion, misinformation, and misidentification. This type of problem often arises because the examinations are conducted by self-trained individuals who lack the required known standard such as handwriting exemplars, fingerprint standards and DNA specimens. Finally, scholarly opinion and connoisseurship, if given by an accepted expert, can often be the deciding factor.

When questions still remain concerning a work's authorship, forensic methods involving a systematic evaluation of the potential trace evidence embedded in the questioned work still remain a valuable, yet untapped, source of data and unbiased proof of its creator.

Dr. Edmond Locard, the founding father of forensic trace evidence analysis, noted in 1929 that whenever a person commits a crime there is always a mutual transfer of trace evidential materials between the people, places, and things involved in the crime. The forensic science literature is filled with case studies where various categories of trace evidence transferred between the persons, places, and objects involved in a crime are used to reconstruct the crime itself. Dr. Locard's principle holds true when an artist creates a work of art.

An artist's intimate contact with his or her work permits the primary, secondary, and tertiary transfer of trace materials between the artist, the environment within which the work is created, and the work itself. Materials such as hairs, fibers, skin cells, fluid droplets, soil, dust, mineral fragments, glass fragments, seeds, plant materials, and other debris can be intentionally and/or inadvertently transferred and subsequently embedded into the work. This can occur in a variety of ways: from the artist to the painting; the painting to the artist; the environment to the artist; the environment to the painting; from the environment to the artist then to the painting; and so on. These tiny traces of particulate matter, hairs, fibers, and fluid droplets can be a valuable source of unbiased, scientific data and proof relating to who created the work, the time period during which the work was created, and where the work was created.

In this study, a painting believed to be the last known work of Jackson Pollock was forensically examined and processed for trace evidence. Items of trace evidence were removed from the painting and compared to materials obtained from the Pollock home and his personal property. Hairs, fibers, and other particles of trace evidence collected from the painting provided unbiased scientific data and proof that the work was produced at the Pollock compound.

Forensic, Fine Arts, Authentication

A93 Computational Methods Supporting Particle Combination Analysis: Application to Very Small Particles on the Surface of Carpet Fibers

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After attending this presentation, attendees will appreciate the powerful support that computational methods provide to the analysis of particle combinations found as dusts on and within virtually any item of evidence. Attendees will understand the distinction, contribution, and requirements of Particle Combination Analysis as compared to more commonly used approaches in

forensic investigations.

This presentation will impact the forensic science community by increasing awareness of Particle Combination Analysis as a new source of forensic investigative information and provide insight into how computational methods assist in the approach. This presentation will also allow program managers and policy makers to recognize the potential contributions of Particle Combination Analysis and to understand the requirements for implementation using existing resources. This will lead to the exploitation of an extraordinarily useful form of physical evidence that is virtually ignored by current forensic science practices.

Particle Combination Analysis is a new approach that uses co-occurring particles to test alternative attribution hypotheses. Simply put, Particle Combination Analysis exploits the particles in dusts, which are ubiquitous and in infinitely varying combinations, to solve a wide range of problems with varying case specifics. This approach can provide a game-changing capability to forensic investigators, working alongside existing investigative methods and using portions of evidence that are typically discarded or ignored.

The approach utilizes existing staff and laboratory resources in a fundamentally different way; it does not require major investments in training, equipment, or retention of outside experts. It does, however, require a change to the way existing resources are used.

An International Criminal Police Organization (INTERPOL) case involving contraband elephant ivory will be used to contrast approaches currently used in most forensic investigations and show how and why a different approach resulted in significantly better results. The same approach can make significant contributions to a wide range of investigations and cases.

Conventional approaches to forensic investigation include: (1) direct comparison to a specific suspected source; (2) classification using a reference library of potential sources; (3) application of a pre-determined analytical tool; and, (4) analysis of specific components of the specimen. Each of these approaches can be useful and makes an important contribution, but each also has requirements that restrict its applicability; none of them fully exploits the available specimen, and their contributions are only serendipitously sufficient to address the needs of any particular investigation.

In contrast, Particle Combination Analysis has no requirement of comparative analysis, no pre-determined restriction on the type of analytical tool, and no pre-determined restriction on the components within the specimen that can be exploited. Rather, standing protocols are used for specimen assessment. The results determine the range of potential contributions to the resolution of case questions. Specific specimen components are analyzed, using modular validated protocols that are not pre-selected, but are strategically chosen based on their potential to resolve case questions.

Using Particle Combination Analysis, dusts from within a shipment of contraband ivory were analyzed to help determine the original location where the ivory was packed. Key findings were the types of minerals, soil, and vegetation represented in the dust, as determined using a combination of light and electron microscopy, energy dispersive X-ray analysis, infrared microspectroscopy, palynology, and non-human DNA analysis. Beginning with a possible origin within the continent of Africa, first-stage analysis of the recovered dusts was able to eliminate environments comprising approximately 91% of the area, including all areas of 36 countries. Of the remaining 12 countries, the analysis was able to eliminate 72% of their area, allowing the investigations to be focused within

portions of these countries. Next steps were defined to further reduce the possible origins of the dust based on more detailed regional analyses.

The Particle Combination Analysis approach resulted in useful information arising from many different particle types. Different combinations of particles resulted in environmental, geographical, and land use “signals” that addressed specific investigative questions in this case. Particle Combination Analysis is a general capability. It is not restricted to questions of geographical origin, to African elephants (or any type of wildlife), to sealed crated shipments, or to specimens containing any specific type of particles. Rather, it is an approach that extracts case-relevant information (for virtually any type of case) from the complex assemblage of particles that are found (on virtually any specimen).

Requirements for successful implementation using existing staff and resources include administrative protocols driving the facile use of multiple disciplines, incorporation of a case-level scientific investigative methodology, and parallel adjustment of institutional norms.

Augmentation of existing practices with this new capability will result in major contributions to many case investigative problems. The primary challenge is institutional willingness to use existing staff and equipment in a different way.

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Particle Combination Analysis, Carpet Fibers, Computational Methods

A94 Evaluation of Within-Roll Variability in Duct Tape Physical Characteristics

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The goal of this presentation is to reveal the within-roll variation present in the following physical characteristics of 55 duct tape rolls: backing color, backing texture, adhesive color, total tape thickness, backing-only thickness, tape width, scrim pattern, scrim count, warp yarn offsets, and the number of backing layers. This presentation is focused on the within-roll variation in the physical attributes of duct tape.

This presentation will impact the forensic science community by improving forensic examiners' evaluations of the significance of physical property differences found within duct tape samples and, as a result, be beneficial for the disassociation of questioned and known duct tape samples.

Research has shown variation in physical and chemical properties between duct tape products, manufacturers, and batches.^{1,2} Although research has shown there are no significant within-roll variations in chemical composition, no data has previously been presented which evaluates the within-roll variation of physical characteristics. The current Scientific Working Group for Materials Analysis (SWGMA) *Guideline for Assessing Physical Characteristics in Forensic Tape Examinations* states, “the analyst must decide what is within an acceptable tolerance” when determining if physical characteristics are consistent between questioned and known duct tape samples.³ No values are provided in terms of expected or acceptable within-roll variability. The lack of research-supported numerical tolerances for these

physical characteristics may have been part of the reason why several respondents failed a recent tape proficiency test in which differences in physical characteristics such as warp yarn offset, width, scrim count, and scrim pattern were cited.⁴

Fifty-five rolls of duct tape, including tape from the four major tape manufacturing companies in North America (Berry® Plastics Corporation, Shurtape®, 3M™, and Intertape Polymer Group®), were obtained. Each was sampled at ten equally-spaced distances down the length of the roll. The 550 samples were randomly numbered to remove examiner bias and the following physical characteristics were then measured or recorded: backing color, backing texture, adhesive color, total tape thickness, backing only thickness, tape width, scrim pattern, scrim count, warp yarn offsets, and the number of backing layers. The variation of total tape thickness, tape width, scrim count, backing only thickness, and warp yarn offsets were statistically evaluated using Microsoft® Excel® software. The possibility of setting tolerance levels with and without uncertainty was evaluated for the disassociation of questioned and known duct tape samples. The following tolerances are recommended based on their ability to include at least 99.5% of within-roll samples without incorporating uncertainty: total thickness $\pm 12\%$, width $\pm 0.15\text{mm}$, scrim count ± 1 , backing only thickness $\pm 35\%$, and warp yarn offset $\pm 0.61\text{mm}$. R statistical software was used to determine if any relationships exist between quantitative and non-quantitative physical characteristics; however, backing texture was the only non-quantitative physical characteristic that had enough representation to evaluate these relationships. No statistically significant relationships were found between backing texture and any of the quantitative measurement variations. This work will improve forensic examiners' evaluation of the significance of physical property differences found within duct tape samples, and as a result, be beneficial for the disassociation of questioned and known duct tape samples.

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Duct Tape, Physical Characteristics, Within-Roll Variability

A95 The Simultaneous Development of Latent Prints on the Adhesive and Non-Adhesive Sides of Tape Using a Rhodamine 6G in Tween 20 Solution After Cyanoacrylate Fuming

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After attending this presentation, attendees will learn of a method to develop latent prints on both adhesive and non-adhesive

sides of tape simultaneously using a solution of Rhodamine 6G, Tween® 20, and water.

This presentation will impact the forensic science community by providing a fast, inexpensive, and simple alternative of processing latent prints on both adhesive and non-adhesive sides of tape simultaneously.

Forensic laboratories process a wide variety of adhesive tapes for latent prints because tapes are commonly used in the binding of victims, the preparation of drug packages, and the manufacture of improvised explosive devices. Even when the tape is touched with latex gloves, latent print evidence can still be recovered. The non-adhesive side of tape is a standard surface for latent print examination with plenty of effective and thoroughly tested development methods, and the adhesive side can be developed with commonly used techniques such as Gentian Violet, TapeGlo™, Sticky-Side Powder™, Wetwop™, and similar powder suspension methods; however, both sides of the tape have to be processed separately, which requires more time and steps for the examiner, and more materials. This study proposes a method by which latent prints can be developed on both adhesive and non-adhesive sides of the tape simultaneously. Strips of a common brand of original-strength duct tape were cut to approximately 15cm in length. Prints from the tips of the thumb, index, middle, and ring fingers of both hands were deposited on both adhesive and non-adhesive sides of the duct tape. The duct tape was then fumed with cyanoacrylate for five minutes, dipped in a solution of Rhodamine 6G, Tween® 20, and de-ionized water for 15 seconds, then rinsed with cool tap water. The prints were visualized using a green (500-525nm) alternative light source and recorded using a digital SLR camera with a 1% orange-red (549nm) long pass filter. The concentrations of Rhodamine 6G, Tween® 20, and de-ionized water were optimized and reliability studies were performed. The optimized solution was then used to process different brands and types of tapes, such as masking tape, electrician's tape, packing tape, etc. Results were compared to standard methods of processing such as TapeGlo™, Wetwop™, and black powder. All results and statistics were calculated by comparing every fingerprint to a custom Likert-type scale where one equals no visibility and five equals excellent visibility. This method results in clear, identifiable prints that are comparable to standard processing methods, yet requires less time and materials.

Latent Print, Adhesive, Tape

A96 Strengthening Research Methodology for New Latent Fingerprints Development Techniques by Means of Design of Experiments (DoE)

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After attending this presentation, attendees will learn how forensic sciences-related research activities can be significantly strengthened and optimized by using Design of Experiments (DoE) and Analysis of Variance (ANOVA) techniques, which represent an efficient procedure based on powerful statistical tools, for planning research experiments so that the obtained data can be analyzed to yield valid and objective conclusions.¹

This presentation will impact the forensic science community by pointing out the importance of accurately planning the experimental activity, which is the basis of every new research.

Although multivariate statistical analysis of experimental data is already widely diffused in several forensic science disciplines, it is usually limited to mere analysis of data obtained by experiments organized by changing one variable at a time. In this way, fundamental relationships and interactions between the factors affecting the process under study can be lost, leading to a relative, not absolute, optimization, which does not always lead to the best solution.

In this framework, DoE can be applied in order to create a predictive model, which is able to describe the relationships existing between the input variables and the responses, which have been selected for a particular process or methodology. Usually, experimental design can be subdivided into the following steps: define the main objective of the experiments; identify the response variables (the dependent ones); select the input variables to be investigated; choose the levels of each input variable; select the appropriate design; perform the experiments; and, analyze the results and create a predictive model.

As far as the choice of the appropriate design is concerned, it strictly depends upon the studied objective, which can be a preliminary screening investigation or an optimization of a known process.

Among the latent fingerprint development techniques, Electrophoretic Deposition (EPD) of inorganic nanoparticles was recently proposed as an innovative and promising procedure to be applied on conductive substrates.^{2,3} Briefly, it consists in the application of an electric field between two electrodes and in the subsequent migration of charged suspended nanoparticles toward the oppositely charged electrode bearing the latent print to be visualized.⁴ The visualization process exploits the inherently present electrical conductivity discontinuities, which are due to the presence of the organic residue.

In this work, this innovative technique was accurate with the main objective of obtaining fully detailed fingerprint patterns. The level of detail produced by the development process and the number of minutiae were considered the responses to be analyzed.

A screening design was first considered in order to establish the most significant variables and the possible interactions among them, affecting the development process, i.e., the responses. Particularly, pH, distance between the electrodes, suspension concentration, and applied potential were considered and varied between two levels. After the screening step, only the key factors were considered in the optimization procedure, for which a response surface methodology was applied.

The presented rigorous systematic research methodology, based on DoE technique, will clearly appear as easily applicable to research activities in several forensic sciences sub-disciplines. Moreover, it fully agrees with the 2009 National Academy of Sciences Report, which recommended strengthening of all the forensic science disciplines. Indeed, this experimental design approach allows evaluating the reproducibility of the studied method and the validity of the chosen model, obtaining from the very beginning full control of the newly undertaken study.

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A97 A Method for the Analysis of Lotions Using Direct Analysis in Real-Time Mass Spectrometry (DART®-MS)

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After attending this presentation, attendees will understand and be able to implement a rapid, simple method for the analysis of complex mixtures such as lotions and the subsequent identification of the components comprising the mixture. Comparison of the results to an analysis using Gas Chromatography/Mass Spectrometry (GC/MS) will be elucidated to evaluate sensitivity and selectivity.

This presentation will impact the forensic science community by providing a validation study for the use of a new methodology in the analysis of forensic samples related to sexual assault investigations. This methodology can result in a more rapid testing and screening of forensic samples to improve laboratory efficiency.

Examination of items of evidence from sexual assaults may, in some instances, require the detection and comparison of materials from lotions and other skin care products used in the commission of the crime. Additionally, understanding the types of components present in lotions will improve the knowledge of background materials expected when analyzing evidence from sexual assault cases to aid in differentiating those types of components from possible condom or sexual lubricant components. Improvements in the ability to detect these types of materials, as well as the ability to rapidly screen items of evidence, will lead to more efficient processing of sexual assault casework.

The validation of a method for the rapid analysis of lotion samples using Direct Analysis In Real Time-Mass Spectrometry (DART®-MS) will be presented. This study included determining reproducibility of the analysis, determining the limit of detection for certain lotion components, blind sampling of unknowns, and the creation of a comprehensible search list. Currently, GC/MS is employed to analyze chemical extracts from lotion samples. While it offers significant specificity, it also requires longer running times. Using DART®-MS, the components of a lotion sample are able to be detected simultaneously using a much shorter analysis time. All components which have been analyzed by GC/MS have been found to be detectable by DART®-MS. Additionally, components that were not detectable by GC/MS were detectable by DART®-MS, demonstrating the increased specificity of this method. The implementation of this method can allow for the rapid screening of multiple samples, which will benefit laboratory efficiency without compromising sensitivity. This technique will prove valuable to forensic science laboratories involved in performing analyses for cases requiring the detection of lotions and their components.

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Lotions, DART®-MS, Sexual Assault Cases

A98 Differentiation of Human Hair by Color and Diameter Using Digital Imaging and Discriminant Analysis

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After attending this presentation, attendees will be introduced to a novel method that offers the potential of providing objective criteria to microscopic hair comparison. The method combines hair diameter with numeric characterizations of red, green, and blue color content determined with the use of digital imaging at defined locations of the hair. Data from different hairs are then compared using discriminant analysis.

This presentation will impact the forensic science community by providing a quantitative means of hair analysis that aims to minimize the subjectivity involved in microscopic hair examinations and strengthen the scientific credibility of conclusions drawn from the comparison of hair samples.

Since hairs are continually shed and are easily removed from an individual, they are a common source of transfer evidence in most types of criminal assaults. Although hair can have important evidentiary value in indicating that contact occurred between individuals, or between an individual and a crime scene, the associative value of hair is often questioned when comparative analysis is based on microscopic comparison, due largely to the subjective nature of the examination.

To assess the viability of this method, ten hairs from each of five participants, all with naturally colored brown hair, were mounted on microscope slides with Meltmount® (refractive index of 1.539) and examined on an Olympus BX53® polarizing light microscope under Kohler illumination. Digital images were viewed with an Olympus DP72® camera at 400x after white balance correction. Using Olympus cellSens Entry® software, the diameter of the hair was measured at the following distance intervals from the base of the root: 500-600um, 600-700um, 700-800um, 800-900um, 900-1,000um, 1,000-1,100um, 1,100-1,200um, 1,200-1,300um, 1,300-1,400um, and 1,400-1,500um. A minimum distance of 500um was selected because previous validation indicated that color measurements begin to show consistency at this point. At each distance interval, a quantitative value for red, green, and blue was determined by the software at three points where the hair diameter was measured. In an additive color model, various combinations of red, green, and blue are used to describe a wide range of colors. The software uses a Red Green Blue (RGB) color model to define each pixel on an image by its principal color components of red, green, and blue. The intensity of each component is provided as an integer ranging from 0-255. Lower values represent less intensity and thus a darker shade of that color component. Therefore, the color gray is described by equal intensities of all three color components relative to each other, while the particular shade of gray is dependent upon how high or low these intensity values are. The intensity of the source was preset to ensure that illumination remained constant prior to the start of hair examination each day during the study. In addition, consistency of the illumination was verified by testing a standard hair to ensure that color values did not fluctuate from day to day.

All diameter and color values for each hair were compared using discriminant analysis (XLSTAT®). Results showed that variation of hairs from each individual was far less than hairs between the five individuals. Two-dimensional analysis showed that hairs from the same individual tended to cluster, easily distinguishing the five individuals. Pairwise comparisons between individuals resulted in p-values significantly less than 0.05 indicating significant differences. These results thus show the potential of this

method to offer subjective analysis of microscopic hair comparison.

Microscopic Hair Examination, Digital Imaging of Hair Color, Discriminant Analysis

A99 Is High Performance Liquid Chromatography (HPLC) a Useful Addition to Current Geo-Forensic Analytical Techniques?

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After this presentation, attendees will be familiar with a number of the different ways in which the results of organic analyses can be used to distinguish between forensically relevant soil samples from close-proximity locations.

This presentation will impact the forensic science community by demonstrating how organic analyses can add evidentiary value to geo-forensic samples and by discussing the results of on-going work into new, practical ways to extract and interpret information from soil, which are independent of, and therefore complementary to, traditional geo-forensic analytical techniques.

Geo-forensic evidence is a potentially useful form of trace evidence for crime reconstruction due to its near ubiquitous presence, high transferability, persistence, and geographic diversity; however, there is a recognized lack of published research assessing the potential of the organic components of this type of evidence. Furthermore, many previous studies have arguably lacked forensic relevance through the use of large sample amounts and the comparison of samples taken from geologically different locations over large spatial scales. Most published geo-forensic studies focus on the inorganic and physical components of soil, which neglects the information contained in the organic fraction of soil, which is dependent on local environmental factors, vegetation, animal matter and anthropogenic contaminants. HPLC is a widely used technique, which has the potential to detect these compounds from small quantities of soil. This presentation will consider whether HPLC is a useful addition to current geo-forensic techniques.

The results of this experimental study will be presented and the application of statistical analysis of HPLC data discussed to illustrate the degree to which it is possible to reliably discriminate between mock crime scene samples taken from close proximity locations of similar geology. In addition, the development of a method for this type of analysis and its optimization, which has led to increased sensitivity and efficiency in comparison with previously reported HPLC methods, will also be presented. The mock crime scene samples were taken from a variety of locations in the United Kingdom, with different underlying geologies in order to illustrate both close proximity and larger distance variability. Temporal variations in the organic composition of the soil, in addition to chromatographic changes caused by variations in sample handling, were monitored and the effects of those changes to sample composition on the ability of the techniques to make useful comparisons were examined. The discriminatory power of HPLC, and its practicality, were assessed by the comparison of the HPLC results and those generated by other independent geo-forensic analytical techniques such as quartz grain surface texture analysis and mineralogical profiling. Lastly, the origin and nature of the diversity of soil chromatographic profiles were also examined

through detailed analysis of major components by methods such as Nuclear Magnetic Resonance (NMR) and Mass Spectrometry (MS).

The results from this study indicate that HPLC can discriminate between locations with up to 100% accuracy and can offer better discrimination than other techniques in the context of samples collected within discrete locations at a small spatial scale. It has also been possible to obtain a four-fold increase in the overall sensitivity of the technique and significantly improve sample preparation and instrument run times, in comparison with previously reported methods, laying the foundation for the greater utilization of this technique in forensic investigations.

Geo-Forensic, Soil, HPLC

A100 Analysis of Fiber Evidence in Motor Vehicle vs. Pedestrian Collisions

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After attending this presentation, attendees will understand the exceptional possibilities for transfer of fiber evidence during motor vehicle vs. pedestrian collisions, the challenges of analyzing such evidence and subsequently associating it to a source, and an example of these techniques applied in a case study.

This presentation will impact the forensic science community by demonstrating one of the key tenets of forensic science, Locard's exchange principle, which states "every contact leaves a trace." Motor vehicle vs. pedestrian collisions are the perfect illustration of Locard's famous theory because they often hinge upon a single fleeting moment of contact between two objects. According to Locard, physical evidence of contact between these two objects always exists, and it is the responsibility and challenge of the forensic scientist to detect that evidence and subsequently to collect it, preserve it, and analyze it.

Presented is a case study of the analysis of fiber evidence in motor vehicle versus pedestrian collisions. In January 2013, an elderly victim using a walker was crossing the street and was struck by a vehicle. The driver fled the scene, and the victim subsequently died due to blunt force impacts to the head, trunk, and extremities, with skeletal, visceral, vascular, and soft tissue injuries. After the passage of 26 days, a black 2010 GMC® Acadia® was recovered and transported to the Cuyahoga County Medical Examiner's Office for examination. Though multiple areas of damage were visible on the front half of the vehicle, no traditional evidence of foreign material transfer (e.g., glass fragments, plastic fragments, paint chips) could be located on the victim's body, the victim's clothing, or the victim's walker that could associate the victim to the vehicle.

Despite the extended period of nearly one month that the vehicle was exposed to the harsh conditions of winter in northeast Ohio, a linear smear of pink material was observed on the hood of the vehicle. Upon close inspection of the victim's pink jacket, a circular area of damage with partially melted edges was located on the posterior left sleeve of the jacket. It was hypothesized that heat generated by the friction and pressure of impact with the hood of the vehicle caused the fibers in the jacket to melt, to fuse together, and to become physically bonded to the substrate. This hypothesis explained both the unusual smeared appearance of the material on the vehicle as well as its persistence over time and in adverse conditions. The apparent fiber material was collected from the smear on the hood of the suspect vehicle, and reference fibers

were obtained from the nylon shell layer and the polyester lining layer of the victim's jacket.

Microscopic and infrared analysis of fiber evidence in which multiple fiber types are fused together presented unique challenges, especially when performing a comparison to known fibers from the victim's jacket. The comparison was complicated by the inability to document traditional fiber characteristics in the smeared material such as diameter, cross-sectional shape, birefringence, etc. The heterogeneity of the smeared material also caused difficulty in determining which areas of the material were purely one fiber type and which areas were a mixture of different fiber types. Fluorescence microscopy proved to be the key to solving both issues.

Utilizing known differences in the fluorescence characteristics (at multiple excitation wavelengths) of the fibers from the victim's jacket, specific areas of the smeared material were identified as pure and/or as mixture. Once these areas were recognized via fluorescence microscopy, they were isolated for infrared analysis, and distinct spectra of both nylon and polyester were acquired within the smeared material. This method allowed for the identification of the fiber types present in the smear as well as an association of the smeared material to the victim's jacket.

It is recommended that scene investigators recognize and vigilantly search for transferred fiber material on vehicles, even when adverse conditions and the passage of time may seem likely to have removed or destroyed such evidence. It is also recommended that forensic scientists become aware of the unique challenges involved in the analysis and comparison of fiber evidence that has been thermally changed by friction and pressure during the moment of deposition onto a substrate.

Fiber Evidence, Microscopy, Fluorescence

A101 Microscopic and Chemical Markers for Cellulose Fiber Combustion

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After attending this presentation, attendees will be aware of microscopically observable markers of cellulose combustion found in residues of burned cotton and other cellulose fabric clothing, and be able to incorporate this information into protocols for collecting fire debris at crime scenes and on the clothing of individuals suspected of setting fires.

This presentation will impact the forensic science community by informing attendees of the findings of microspheres and sticky amber residues in burned debris, and finding levoglucosan in the amber residues, which can provide markers of cellulose fabric burns and exposure to the fire. This can be applied to forensic science casework by establishing types of clothing burned in burn barrels or burn pits to destroy evidence, and as markers on the clothing of a person exposed to a fire in its early stages.

When cotton and other cellulosic fabrics are burnt or exposed to heat, the pyrolysis products include char, ash, mineral residues including light-colored microspheres, black hollow microspheroids, and an amber-to-brown viscous material. The viscous amber material was previously reported and identified it as levoglucosan (1,6-anhydro- β -D-glucose), a known pyrolysis product of cellulose combustion. This presentation expands the identification and demonstrates the genesis and behavior of the amber droplets and their relationship to the black microspheres.

Fabrics other than cotton (rayon and linen) also produce the amber droplets. Sources of levoglucosan other than cellulose were tested, as were amber droplets that did not contain levoglucosan. Residues of fireplace burns and a beach bonfire for field evidence were examined.

Levoglucosan, a white crystalline solid when pure, is encountered as a major component of sticky amber and brownish residues on heat-damaged cellulosic fabric items, and as microscopic droplets that are deposited on protected areas of the fabric. In test burns conducted with the use of temperature probes, such areas, protected by folds in the fabric, remained at or near ambient temperature for at least 30 minutes after ignition, although the surrounding fabric quickly rose to over 800°F. The amber residue forms as an aerosol, resulting in droplets where it condenses, and as sticky residues where the droplets collapse or are dissolved in water — another product of cellulose combustion. As once-protected areas burn, the amber droplets undergo secondary pyrolysis, forming hollow black microspheres. The production of levoglucosan in test burns and its identification by Liquid Chromatography/Mass Spectrometry (LC/MS), Gas Chromatography/Mass Spectrometry (GC/MS) and pyrolysis-GC/MS is described. The study explored use of levoglucosan as a marker for cellulose burns in a forensic science context and report microscopic traces of amber residue — shown to contain levoglucosan — on tapelifts of cotton shirts “worn” by manikins placed in proximity to a test burn pile of cotton garments.

No previous reports of black microspheres resulting from cellulose combustion were found, and no reports in the forensic literature, textile science literature, or textile fabric flammability studies of the amber droplets that were observed. However, the formation of levoglucosan and other products of cellulose combustion is well-established, elucidated in early studies of cellulose pyrolysis by Shafizadeh et. al.^{1,2} Aerosols (i.e., sub-microscopic droplets) of levoglucosan are reported in the atmospheric science literature as indicators of forest burns. Textile fabric flammability studies indicate levoglucosan to be the initial low-temperature product that readily supports flaming combustion. Maximum levoglucosan yield is reported at 250°C, and secondary pyrolysis is reported to begin above 240°C. Studies of waste materials as a fuel source report levoglucosan as one of the products that undergoes further pyrolysis. Levoglucosan has been identified in archaeological samples of char from ancient burn pits and is now considered a marker of cellulose combustion; it is stable and unique in this context to cellulose pyrolysis. Reports of test burns using contemporary woods and rice straw buttress this research. Atmospheric samples of aerosols consisting of microscopic droplets from agricultural burns also yield levoglucosan, which has been proposed as a marker for such events as well as for forest burns. This literature dovetails with the experimental determination of the aerosol source of amber droplets from test burns of cotton blue jeans and t-shirts, and its applications to forensic science questions.

The black microspheres and amber droplets resulting from the test burns are much larger than atmospheric samples, being visible using a stereobinocular microscope at magnifications as low as 16x. They can be detected during routine preliminary examination using techniques that allow for rapid decisions about sampling and further testing. For example, debris from a burn can be sieved and black microspheres found in the smaller fractions. Amber droplets or sticky residues can be found on surfaces of fabrics and items that may have been nearby.

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Thermal Damage, Fire Debris, Levoglucosan

A102 Laboratory and Field Markers of Cellulose Fabric Pyrolysis and Recommendations for Collection and Preliminary Examination of Burn Residues

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After attending this presentation, attendees will be aware of the factors affecting the forensic use of levoglucosan as a marker for cellulose fabric combustion and can make decisions about selecting microscopically observed residues for further testing, and have an overall view of finding and analyzing samples in both field and laboratory.

This presentation will impact the forensic science community by showing that findings of levoglucosan-containing droplets, sticky amber residues from their collapse, and black microspheres from their secondary pyrolysis can be used to assist with temperature mapping at fire scenes, as they are markers of cellulose fabric combustion with specific temperature thresholds.

Amber droplets produced during test burns of cotton blue jeans and t-shirts contain levoglucosan (1,6-anhydro- β -D-glucose), a known product of cellulose pyrolysis, considered to be a marker of cellulose combustion in atmospheric science, archaeology, and studies of agricultural burns. Levoglucosan droplets, sticky residues from their collapse, and the black microspheres that result from their secondary pyrolysis have previously been reported. Such residues can also be produced by burning ordinary table sugar and the many foods and beverages in which sugar is a principal component. Levoglucosan aerosols were produced from sugary materials by heating them in a crucible and collecting the resultant aerosol on microscope slides. Moreover, there are numerous sources of sticky amber and brownish residues that do not contain levoglucosan — amber droplets were produced with no detectable levoglucosan from piñon tree incense. The presence of levoglucosan can be established only with testing such as Liquid Chromatography/Mass Spectrometry (LC/MS), and Gas Chromatography/Mass Spectrometry (GC/MS).

It would be useful to have a quick preliminary test to determine whether sticky amber residues warrant further testing. Filtering polar extracts of the amber residue through activated charcoal and glass wool does clean up the extract, but not completely. Levoglucosan recrystallized from solution, but not rapidly enough to be useful in screening; however, the solubility of the amber residue in water and acetone distinguishes it from most other types of amber sticky residues. This screening test can be performed with minimal sample by touching a dissecting needle to an amber residue then touching the needle to water and acetone on a microscope slide. Sugary residues are also water-soluble, but the solid residue from sucrose test burns did not include structures such as burnt fibers.

In order to evaluate field residues in natural conditions, residues from beach bonfires and fireplace residues from burns using wood and pressed logs were sieved and examined. The fireplace residues included tiny black, white, and light-colored

microspheres, as well as particles unlike those from cotton burns, such as burnt wood cells and blackened microspheres that exhibit cell structure. No sticky residues were observed, but no extractions were attempted. Microspheres were not found in the beach bonfire sample, perhaps because they were washed away. No extractions or examinations at higher magnifications than afforded by a stereobinocular microscope were performed.

The temperatures at which amber levoglucosan droplets first appear and then undergo secondary pyrolysis to black microspheres, together with the size of the droplets, may provide information about heat travel at fire scenes. It would be useful to determine how far levoglucosan-containing droplets travel, how distance is affected by the size of the droplets, and how droplet size correlates with burn conditions. Hopefully, research will be stimulated in this area.

Some suggestions: (1) if fabric weave structure is visible in burned residues, collect this intact; do not sieve or sort at the scene until after these are collected; (2) when sieving the remaining burn residue at a scene, save the larger artifacts, but also smaller fractions, as they may include smaller artifacts such as zipper teeth and so on; (3) analyze burnt fabric for fiber content and weave; portions of the weave often survive intact, as do some fibers. The original identity of more severely burnt fibers may be inferred from the pyrolysis products; (4) examine delicate ash that preserves fabric structure; (5) after removing larger fragments, sieve the debris using fine-mesh sieves. Debris may include microspheres typical of cellulose burns, including black microspheres, white microspheres of carbonate, and hardened droplets of synthetic melt from trim, thread, zippers, etc.; (6) examine burnt fabric items under a stereomicroscope and look for amber droplets and black microspheres adhering to the fabric; (7) when examining the clothing of a person suspected of being near a fire, look for amber droplets and black microspheres as well as amber-to-brown sticky residues, as these may contain levoglucosan or its pyrolysis products; (8) if no evidence of exposure to heat is initially observed, but is suspected, take tape lifts of those areas of clothing items most likely to have been exposed to heat; and, (9) sticky amber residues that dissolve in water and acetone are good candidates for further testing using LC/MS or GC/MS.¹

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Thermal Damage, Fire Debris, Levoglucosan

A103 Systematic Study and Analysis of Thermal Decomposition Products Generated From the Combustion of Porcine and Human Tissue Under Laboratory Conditions

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The goal of this presentation is to ensure that attendees will: (1) gain a better understanding of the process of generating thermal decomposition products from porcine and human tissues; (2) understand the type of products generated from the pyrolysis of these samples; (3) compare and contrast the type of products generated across these two samples and deduce whether porcine samples are a good experimental model for human tissue in such studies; (4) identify potential characteristic volatile indicators of human remains in a fire; and, (5) investigate the effects of

decomposition on the type and abundance of products generated from human samples across a 20-day decomposition period.

This presentation will impact the forensic science community, especially in the field of fire investigation, as the work explores the pyrolysis products from cadaveric samples. A systematic study to generate, analyze, and identify thermal decomposition and pyrolysis products produced from burning porcine and human samples at elevated temperatures under laboratory conditions is described. This presentation contributes in validating the accuracy of the use of porcine samples as a substitute of human samples in laboratory-generated pyrolysis data and it also reports key chemical indicators of cadaveric remains within fire debris samples that will potentially aid investigators dealing with such samples.

When organic materials degrade into smaller compounds at elevated temperatures (typically in a fire) in the absence of oxygen, thermal decomposition or pyrolysis occurs.^{1,2} Products generated from this process can act as background interference products in the analysis of fire debris for Ignitable Liquid Residue (ILR).^{3,4} While numerous studies have been conducted in identifying such interfering products from non-biological sources, it is only in recent years that attention has focused in the direction of identifying volatile products from burned porcine and human remains.⁵⁻⁹ When a human body is subjected to heat, this complex fuel package can dynamically contribute in fuelling a fire, given the right circumstances.^{8,10}

Focusing on developing a systematic study for volatile data generated under laboratory conditions, a methodology was developed whereby porcine and human samples were burned under laboratory-controlled conditions and the volatiles generated adsorbed onto Activated Carbon Strips (ACS), desorbed in solvent, and analyzed and detected using Gas Chromatography/Mass Spectrometry (GC/MS). Products generated from porcine and human samples were compared and observed to have similar pyrolytic profiles containing predominantly n-alkane, n-alkene, aromatic compounds (benzene, toluene, p-xylene, alkylbenzene), and nitriles. Alkylcycloalkanes and n-aldehyde were also present consistently in porcine samples but not detected in any of the human samples. Similar outcomes were noted for some of the heavier n-alkanes and n-alkenes.

This study also examined the changes in composition and abundance of pyrolysis products generated over time from decomposing human samples across a 20-day period under laboratory conditions. Although the results suggested that the composition of these products remained more or less the same across the experimental period, a patterned rise in the abundance of products was observed from day three peaking at day six before decreasing at day 20 across six repeated tests.

The methodology developed in this study proved to be robust with acceptable reproducibility attained across the sampling process. The results also indicated that human and porcine samples produce relatively similar pyrolysis profiles with only minor differences, suggesting that porcine samples are a good representative for human samples.

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Thermal Decomposition, Cadaveric Material, Pyrolysis Products

A104 Development of an Expert System for Classifying Ignitable Liquid Residues in Fire Debris by ASTM E1618

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The goal of this presentation is to acquaint the user with a new expert system developed for classifying Gas Chromatography/Mass Spectrometry (GC/MS) data from fire debris analyses into one of the various ignitable liquid classes according to the American Society for Testing and Materials (ASTM) E1618 standard method. This will help reduce the subjective nature of manual assignment by the analyst and associate a "match" index for the assignment.

This presentation will impact the forensic science community by providing improved E1618 assignment as well as a more objective method for quality of match.

The ASTM E1618 method establishes criteria for the fire debris analyst to classify ignitable liquid residues into one of eight different classes based on visual inspection of extracted ion chromatograms from GC/MS data. Some cases require identification of specific compounds based on mass spectra. An expert system was developed which predicts which class based on the same extracted ion chromatograms. The data set used for this expert system is the complete Ignitable Liquid Reference Collection (ILRC) maintained by the National Center of Forensic Science (University of Central Florida) and Technical Working Group for Fire and Explosives (TWGFEX) of 548 commercial products representing all classes under E1618, which either may be used as an accelerant in commission of arson or be incidental liquids found in fire debris. Like mass spectral library search programs, a similarity index is provided as a measure of confidence in the classification. Also like any search or matching algorithm, the analyst should confirm the match by his/her own inspection of the data.

Classification from preliminary tests give correct assignment greater than 95% (lowest for oxygenate and miscellaneous classes) and the correct classification was listed in

the top three picks for IRLC samples. E1618 standard recommends identification of specific compounds based on mass spectral matches for these classes, especially when only a few compounds are present. The expert system works with only extracted ion data rather than full mass spectral data. These classes as well as samples resulting in a low match are flagged for analyst attention and verification of assignment.

Aside from inclusion of an internal standard (3-phenyl toluene) in the carbon disulfide solvent used in the activated charcoal method (ASTM E1412), no special columns or temperature programs are required. The extracted ion chromatograms are exported from GC/MS software as Comma Separated Variables (CSV) and imported into the expert system, and thus is vendor independent. The retention time and area counts of the internal standard are added as the first line in the data file. The internal standard is used to determine relative retention times of individual components and is only a semi-quantitative measure of the concentration of individual compounds. Data from an ASTM E1618 standard mix is used to determine the "high," "medium," or "low" subclasses of each E1618 class.

The expert system and data processing software are written in the free open source statistical software language R (<http://www.r-project.org/>). The expert system was tested using actual fire debris samples where the ignitable liquid known was independent of the training data set or as diluted liquids simulating evidence collected from a container. Effect of substrate on classification will be presented.

ASTM E1618, Ignitable Liquids, Expert System

A105 Hierarchical Cluster Analysis of Ignitable Liquids

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After attending this presentation, attendees will appreciate the utilization of the Total Ion Spectrum (TIS) for statistical classification methods of ignitable liquids since results from hierarchical cluster analysis produced a comparable classification scheme to the American Society for Testing and Materials (ASTM) E1618-11.

This presentation will impact the forensic science community by substantiating the ASTM E1618-11 classification scheme. The goal of this research is to establish the effectiveness of the TIS for statistical classification methods that can provide known error rates for classification of ignitable liquids.

Classification of ignitable liquids for fire debris analysis is performed following the ASTM E1618-11 classification scheme. Seven major classes are employed: aromatic products; gasoline; petroleum distillates; isoparaffinic products; naphthenic-paraffinic products; normal alkane products; and, oxygenated solvents. Ignitable liquids are assigned to a miscellaneous category if they do not meet the criteria of one of the seven classes. The seven major classes, except for gasoline, are further sub-classified based on the n-alkane range of the ignitable liquid into light ($C_4 - C_9$), medium ($C_8 - C_{13}$), and heavy ($C_9 - C_{20+}$). Assignment of an ignitable liquid into an ASTM class is determined by the analyst's interpretation of the Total Ion Chromatogram (TIC), Extracted Ion Chromatograms (EIC), and the presence of target compounds.

Hierarchical cluster analysis is an unsupervised learning method for grouping samples without having any prior knowledge

of group membership.¹ The process begins by considering each sample individually, then sequentially grouping the most similar samples, and lastly combining similar groups to form new groups. Ideally, samples within a group should be more similar to one another than to samples in other groups.² Results are typically shown as a dendrogram, which displays how the samples or groups are related by using connecting lines whose lengths reflect the differences between them.

The National Center for Forensic Science (University of Central Florida), in collaboration with the Scientific Working Group for Fire and Explosions (SWGEX), has developed and maintains an ignitable liquids reference collection and database.³ The GC/MS data of 445 ignitable liquids were employed in this research. ASTM E1618 classification assignments were performed by a SWGEX committee of practicing fire debris analysts. Hierarchical cluster analysis was performed with the ignitable liquid TIS obtained from the GC/MS data. The TIS is equivalent to the average mass spectrum across the chromatographic profile.

Results from hierarchical cluster analysis of the TIS-grouped ignitable liquids are based on their chemical characteristics. Most ignitable liquids grouped together by hierarchical cluster analysis were also of the same ASTM E1618 class as designated in the Ignitable Liquids Reference Collection database.⁴ These results support the use of the TIS in statistical methods of classifying ignitable liquids.

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Fire Debris, Ignitable Liquids, Hierarchical Cluster Analysis

A106 Fire Debris: Research and Database Initiatives at NCFS

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After attending this presentation, attendees will understand the synergistic efforts in fire debris analysis research and ignitable liquids database development at the National Center for Forensic Science (NCFS).

This presentation will impact the forensic science community by providing fire debris analysts with resources for casework and training through exposure to an overview of results from peer-reviewed research and ongoing database efforts.

Previous research in the area of fire debris analysis at NCFS has shown that the average mass spectrum across the chromatographic profile can serve as a retention time— independent basis for classifying ignitable liquids, comparing data from different laboratories, and rapidly searching libraries of ignitable liquid data.¹ The average mass spectrum across the chromatographic profile has been referred to as the Total Ion Spectrum (TIS) and has also been shown to be useful in identifying an ignitable liquid signal in a strong pyrolysis background through a combination of target factor analysis and Bayesian decision theory, resulting in a unique soft classification system.² In more recent research, the TIS has also been shown to be useful in classifying fire debris residue as positive or negative for ignitable liquid residue using linear or quadratic discriminant analysis and Soft Independent Modeling of Class Analogy (SIMCA).^{3,4} In more recent work, the TIS have also been shown to be useful in the identification of the ASTM E1618 classes of ignitable liquids using Self Organizing Feature Maps (SOFM).⁵ An overview of the results from these research studies will be given.

Research in the area of fire debris analysis at NCFS has been significantly aided by the access to ignitable liquids data from the Ignitable Liquids Reference Collection Database (ILRC) and Substrates Database. The ILRC contains records on over 600 ignitable liquids that have been analyzed under a standard protocol and assigned to ASTM E1618 class designations by a group of practicing fire debris analysts in the Scientific Working Group for Fire and Explosions (SWGEX). These databases contain only records on ignitable liquids and substrates purchased in the United States; however, they serve as a resource for casework and training, both within the U.S. and abroad. In an effort to broaden the representation of ignitable liquids in NCFS databases to include samples from around the world, an International Ignitable Liquids Database has been initiated. The NCFS is working with the European Network of Forensic Science Institutes' (ENFSI) Committee on Fire and Explosions to begin populating the database. As this project moves forward, other fire debris analysis organizations from around the world will be invited to partner in the development of this international resource. A brief overview of these databases will also be provided.

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Fire Debris, Research, Database

A107 Development of Paper Microfluidic Devices for the On-Site Detection of Improvised Explosives

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The goal of this presentation is to show the development of paper microfluidic devices for the rapid, on-site detection of improvised explosives composed of items such as black powder, flash powder, Ammonium Nitrate/Fuel Oil (ANFO), urea nitrate, and military explosives. The development and design of these devices as well as the limits of this method will be discussed.

This presentation will impact the forensic science community by showing that as explosives become easier to manufacture and terrorism increases, the demand for rapid, on-site detection of explosives is also increasing. Paper microfluidic devices are inexpensive, stamp-sized devices that can be designed with multiplexed colorimetric tests to detect the presence of a range of explosive components. These chips permit multiple compounds to be detected on a single device in order to obtain a presumptive identification of the material before it is sent to a laboratory.

In recent years, there has been a dramatic increase in the use of improvised devices, in part due to better controls placed on commercial and military explosives. In addition, a large number of procedures describing methods to prepare improvised explosives have been posted on the internet, making it easier for terrorist and amateur bomb makers to build them. Therefore, police and forensic evidence collection teams need a rapid, inexpensive, simple method in order to identify these types of materials in the field.

Commonly used materials for this type of explosive preparation include fertilizers and industrial chemicals containing oxidizers such as chlorates, perchlorates, and nitrates as well as other less stable compounds, such as peroxides. Because these materials have a wide range of volatility, polarity, and composition, multiple analyses must be run in a lab in order to identify the explosive material. Gas Chromatography/Mass Spectrometry (GC/MS) and Liquid Chromatography/Mass Spectrometry (LC/MS) can be used to identify organic compounds while Ion Chromatography (IC) and Capillary Electrophoresis (CE) can be used to identify inorganic ions. Metals can be detected using Scanning Electron Microscopy/Elemental Diffraction Spectroscopy (SEM/EDS) or X-Ray Diffraction (XRD). These expensive pieces of instrumentation are not portable, requiring the sample to be sent to a laboratory for testing. This process increases the amount of time before the identity of the explosive can be provided to on-site personnel.

The use of paper microfluidics allows for the development of very inexpensive analytical devices based on designs printed in wax-based ink on chromatography paper. These wax-designed channels can direct liquid samples toward individual sections of the paper containing colorimetric test reagents. Based on the design, a simple device can perform five or more simultaneous analyses while costing very little, since the basic design components (paper, wax, and small quantities of reagents) are all very inexpensive. These devices can also be easily stored for long-term performance and, due to their low cost, could revolutionize on-site forensic testing.

These devices will be capable of detecting nitrate, nitrites, peroxides, urea nitrate, ammonium, chlorate, perchlorate, chloride, sugar, cyclonite, trinitrotoluene, zinc, aluminum, magnesium, and sulfate. This allows for the design of three different chips for the

detection of multiple components in an improvised explosive, such as flash powders and other low explosives, along with post-blast identification capabilities.

The development of this paper-based sensor will allow police and forensic evidence collection teams a simple, reliable device for the presumptive testing of unknown evidence in the field. These paper devices can be easily stored and take up very little space, allowing them to be easily brought into the field with the team.

This project was supported by NIJ Award #2012-DN-BX-K048 awarded by the National Institute of Justice, Office of Justice Programs, U.S. Department of Justice. The opinions, findings, and conclusions or recommendations expressed in this presentation are those of the author(s) and do not necessarily reflect those of the Department of Justice.

Explosives, Paper Microfluidics, On-Site Detection

A108 Forlab Concept: Forensic Laboratory for *In Situ* Evidence Analysis in a Post-Blast Scenario

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After attending this presentation, attendees will learn a different approach to the crime scene search after an Improvised Explosive Device (IED) explosion, which involves multidisciplinary screening techniques, specifically designed to increase the efficiency of the collection of the items.

This presentation will impact the forensic science community by introducing a new idea to approach the collection of pieces of evidence in a post-blast scenario, which uses on-site contactless techniques in order to sensitively reduce the number of objects seized at the scene of the event, with the ultimate goal of increasing the efficiency of the forensic analysis.

An IED post-blast scenario can cover a vast area where massive and diverse pieces of evidence must be found, collected, and transferred for analysis to a reference laboratory, often based in a distant place from the scene of event. The evidence collection is undertaken without any feedback from the data repository; thus, large volumes of unnecessary analytical data are generated in the forensic laboratory due to the analysis of items not relevant for the investigation. These complex tasks involve tremendous amounts of material and human resources. Forlab will deliver a novel systematic methodology for optimizing the evidence collection by maximizing the speed, the reliability, and the accuracy of the collection process.

The concept of Forlab is to assemble a technology that allows the end user to have at the same time the on-site detection, identification, and analysis of evidence, the production of a 3D recreation of the scene, and deliver in real time all the relevant information of the scene readily available in a delocalized Command and Control Center (CCC) to the expert leading the forensic investigation.

Through the analysis of the data coming from the scene of event, the CCC can guide the crime scene officials, providing them the necessary information to select the seized items properly. The objective is to optimize the evidence collection and to reduce the time and the resources in the laboratory while preserving the chain of custody so as to minimize the time required to identify those responsible for the attack.

The proposed system consists of analytical technologies for *in situ* sample screening, communication, and positioning modules for localizing the information and transporting it further and providing the CCC with 3D scene recreation capabilities.

The selected technologies to develop the portable

sensors are: (1) Laser Induced Fluorescence (LIF), a standoff technique to acquire fluorescence maps to detect different types of evidence (e.g., plastics, textile fibers, and polymers, in general) in the scanned area; (2) Laser Induced Breakdown Spectroscopy (LIBS) coupled with Raman spectroscopy, a portable device to obtain chemical information from the collected evidence and other materials not easily removable from the scene of event, in order to detect traces of explosives and other chemical elements; and, (3) Non-Linear Junction Detection (NLJD), a standoff technology to detect electronic debris, even those not visible to the naked eye.

The forensic science community must have a real benefit in terms of logistics, analytical procedures, personnel education level, training, and competency if such an approach could produce a ready-to-use tool to support the investigation in the crime scene in a post-blast event.

Improvised Explosive Devices, On-Site Trace Analysis, Crime Scene Search

A109 An Evaluation of Pseudo-Explosive Training Aids

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After attending this presentation, attendees will be better informed as to the nature and efficacy of pseudo-explosive training aids, a proposed alternative to genuine explosives in the training of bomb-sniffing dogs.

This presentation will impact the forensic science community by serving to evaluate existing commercial pseudo-explosives. It will explore whether pseudo-explosives are a valid substitute for genuine explosives and whether or not they are dependable enough to come into general use.

It is widely recognized that canines are the fastest, most sensitive, and selective real-time detectors of explosives that are currently available. The rearing, training, and upkeep of explosives-detecting canines, while not trivial in cost, is still cost-competitive with specialized instrumentation designed for on-site explosive detection. What is essential to maintaining a properly trained explosive-detecting canine, however, are training aids that are uncontaminated and representative of the various explosive classes that exist. Such training aids have traditionally consisted of samples of authentic explosive formulations. This introduces challenges of proper acquisition, storage and destruction of the explosives, as well as restrictions on locations in which actual explosives can be deployed for training (e.g., airports and airplanes). Hence, it has been suggested that "pseudo-explosives," which simulate the odor of actual explosives yet contain only inert ingredients, should be used in the training and testing of explosive-detecting canines. An obvious concern with this approach is the unknown chemical composition of pseudo-training aids. Furthermore, it has not been established in testing by an independent party that canines trained on pseudo-explosives will successfully detect actual explosives. Similarly, it also must be established that canines trained on an actual explosive respond to the pseudo version of that explosive. Hence, this project was carried out in two phases: chemical analysis of pseudo-explosive training aids, as well as canine trials to determine their effectiveness. In particular, two different brands of pseudo-explosive products were evaluated: the Controlled Odor Permeation System (COMPS), which is manufactured by Florida International University, and ScentLogix™-K9 training aids. In each case, simulants of TNT, Composition C-4, and smokeless powder were used along with the actual explosives. It was established that

these aids largely consisted of pure COMPS bags or silica particles that were coated with a mixture of odor compounds (ScentLogix™). The odor compounds themselves were classified as either “Explosive Related Compounds,” which are energetic impurities or degradation products of an actual explosive, or “Signature Compounds,” which are non-energetic solvents, stabilizers, and other ingredients of explosive formulations. Canine testing was used to determine whether canines trained solely on pseudo-explosives can detect authentic explosive samples as well as whether canines trained on explosives will respond to the pseudo-explosives. In these trials, canines trained on pseudo-explosives were generally unable to locate the actual explosives that were being simulated by the training aid. In addition, canines trained on actual explosives showed little to no interest in the pseudo products.

Pseudo-Explosives, Canine Training Aids, Explosives-Detecting Canines

A110 What’s in a Name? That Which We Call “Touch DNA” Perhaps Should Be Split in Twain

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After attending this presentation, attendees will have a clearer understanding of the nuances of evidence triage in an era of tests which are more sensitive than ever.

This presentation will impact the forensic science community by generating discussion and introducing proposed terminology for “classifying” samples containing DNA primarily from sources other than the classical DNA-rich fluids of blood, semen, or saliva.

The term “touch DNA” has entered the field of forensic biology. Early on, it was used to describe the small amount of DNA found in a fingerprint, then became conflated with Low Copy Number (LCN) testing, and has further expanded in some usage to mean any sample whose DNA comes from anything other than body fluids. The use of the term has caused confusion and disagreement amongst many forensic stakeholders, such as police agencies, lawyers, and judges. The term is now pervasive enough that it has its own entry in Wikipedia®.

In news articles, popular science writing, and even in scientific forums, “touch DNA” is being used in a broad and inconsistent manner. This study proposes that a more defined and precise usage be substituted: those categories of samples that tend to have enough DNA for typing without resorting to enhanced LCN techniques could be called “handled items” and those that tend to require those enhanced LCN techniques to obtain a DNA profile would retain the “touched items” nomenclature.

Data collected by the Department of Forensic Biology over the last several years was used to evaluate different types of evidence commonly received. The information included sample type, yield of DNA, quality of DNA profile, and suitability for entry into CODIS. The data will be presented along with suggestions for classification of sample types into either the “handled” or “touched” category.

In any laboratory, such information can be used to: (1) develop case acceptance criteria; (2) guide the “triage” of items within larger cases; or, (3) determine the analytical path an item will take. Examples will be shown of decisions made by the Department of Forensic Biology with regard to all three.

Using the two terms with precision can lead to increased

clarity when speaking with those who collect the evidence or present the results in court. With a better understanding of the nature of the evidence item, expectations of success would be appropriate for each category, leading to better satisfied customers.

Touch DNA, Evidence Triage, Handled DNA

A111 Recovery of Touch DNA From Pipe Bombs and Their Containers Following Various Rendering-Safe Techniques

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After attending this presentation, attendees will learn about the strategies used to obtain touch DNA evidence from various explosive devices and/or proximal containers following two “rendered-safe” techniques.

This presentation will impact the forensic science community by introducing the new touch DNA methods for identifying individuals associated with the assembly and/or handling of explosive devices following common strategies for safe rendering. Furthermore, it will assist bomb squads in knowing which rendering-safe technique to use on improvised explosive devices and their proximal containers if the aim is to optimize the yield of DNA recovered.

Crimes involving explosive devices have been on the rise in both domestic and foreign attacks. Explosives are involved in approximately 70% of terrorist events. While the types of improvised explosives vary, pipe bombs are common explosive devices that make up approximately 30% of these attacks. Most recently, the two Boston Marathon bombing suspects utilized a multitude of explosive devices including pipe bombs, grenades, and improvised explosive devices. Although these bombs vary in strength and sophistication, all required manual assembly using common materials.

The ideal outcome of a bombing attempt is neutralization of the explosive device by a bomb disposal unit. There are different techniques used to render a pipe bomb safe, all of which involve opening the device in some manner to reduce pressure and prevent explosion. Until recently, pipe bombs were rendered safe by firing a high-velocity clay projectile at the midpoint of the explosive. A relatively new technique using high-pressure water in place of clay has been developed. In the current study, these two techniques were compared to determine their impact on recoverable DNA left on the surface of the pipe bomb and its proximal container.

Thirty known subjects were asked to wear and handle a backpack for seven days. Touch-DNA samples were collected from each backpack using laboratory developed methods. Polyvinyl Chloride (PVC) pipes were used to create mock pipe bombs with non-explosive materials. Pipe bombs were placed inside each backpack and were later rendered safe by the following methods: ten were rendered safe with the use of a clay projectile; ten were rendered safe with the use of a high-pressure water technique; and, ten were sampled without being rendered safe (control samples). The end caps of the bombs, backpack back straps, and backpack zippers were swabbed for DNA analysis. Each sample was extracted, quantified, and analyzed using Short Tandem Repeat (STR) analysis.

Due to a small sample size, no statistically significant difference was found in DNA quantitative recovery as a function of the rendering-safe technique. However, preliminary quantitative

data do indicate a trend favoring the clay method with respect to DNA analyses. A difference in terms of success rate of obtaining DNA profiles as a function of rendering-safe technique was observed. While only 50% of pipe bomb end caps that were rendered safe with water produced partial profiles and 10% produced full profiles, 90% of pipe bomb end caps rendered safe with clay produced partial STR profiles. Work to increase sample size and DNA recovery from backpacks is currently in progress. Non-rendered safe controls demonstrated higher DNA concentrations than those rendered safe. This study demonstrates that while rendering safe techniques are necessary for ensuring safety, the approach used can have serious consequences in a case with respect to the potential for identifying a suspect.

STR Typing, Pipe Bomb, PowerPlex® 16HS

A112 Optimizing Techniques for DNA Recovery and Extraction From Spent Cartridge Casings

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After attending this presentation, attendees will understand the enhancement of methods for the retrieval and extraction of DNA from spent cartridge casings commonly recovered at crime scenes.

This presentation will impact the forensic science community by introducing improved techniques for recovering and processing trace DNA from spent cartridge casings in order to obtain the highest DNA yields, along with the most genetic data. The findings have the potential to influence the procedures DNA analysts utilize when recovering DNA from spent casings and ultimately identifying the individual who handled casings recovered from a crime scene.

In an incident where a firearm is discharged, it is critical that investigators have access to reliable forensic tools capable of successfully identifying the person(s) responsible for firing that weapon; however, the best evidence for a shooting incident — the actual weapon used — is rarely recovered from a scene. What are often recovered are cartridge casings ejected from a firearm, which have the potential to provide direct links between the incident, the weapon, and the perpetrator.

Fingerprints may be transferred to cartridges when the weapon is loaded; however, retrieving them following firing is rarely successful. As a result, some crime laboratories have attempted to recover and analyze DNA left on spent casings via autosomal Short Tandem Repeat (STR) analysis. In general, these efforts involve swabbing the casings and utilizing the laboratory's Standard Operating Procedure (SOP) for processing swabs. Limited attempts have been made to overcome the low copy, degraded state of the DNA on this type of item. Owing to this, few laboratories have encountered STR typing success from DNA on spent casings. Thus, it is essential that methods for DNA retrieval and analysis be optimized for the highly compromised touch DNA that exists on them. In the research presented here, multiple variables, such as: (1) swabbing versus soaking casings; (2) shaking the casings during soaking; (3) pre-incubation at 85°C; (4) shaking swabs during digestion; (5) the duration of digestion; and, (6) pretreatment of purification columns were examined with the goal of improving DNA yields and typing results from touched casings.

Soaking methods were evaluated so as to minimize soak volume. The bulbs of transfer pipettes are inexpensive and disposable, with diameters just larger than that of casings, which proved ideal for soaking. The bulbs, along with live cartridges,

showed no contamination with exogenous DNA. Cleaned cartridge casings were handled by volunteers and DNA yields from swabbing and soaking, along with the other variables listed above, were compared. Three extraction methods — a silica-based extraction kit (Qiagen® QIAamp® DNA Investigator Kit); a sorbent-based extraction kit (nexttec™ Biotechnologie GmbH Fingerprint DNA Finder (FDF) Kit); and a standard organic extraction — were then tested to determine which was most successful in maximizing DNA yields and STR allele recovery.

Following optimizations, volunteers loaded pistol magazines with five cartridges, which were fired and casings collected. Five DNA recovery and purification procedures were compared: (1) double swabbing and organic extraction; (2) soaking and organic extraction; (3) double swabbing and a silica-based extraction; (4) soaking and a silica-based extraction; and, (5) single swabbing followed by a sorbent-based extraction. DNA yields were quantified using real-time PCR, and STRs were assayed using the Life Technologies® MiniFiler™ PCR Amplification Kit. Statistical analysis of DNA yields, STR profile types (percent of full profile recovered), and allelic consistency with the handler were calculated.

Double swabbing and organic extraction resulted in the highest DNA yields, generating up to 90% or more of alleles consistent with the handler. Soaking and organic extraction performed almost as well; however, these two methods also resulted in the most allelic drop-in. In contrast, the sorbent-based extraction, which was specifically designed for touch samples, only generated 25% or fewer alleles consistent with the handler, but little or no drop-in was observed. Finally, the silica-based protocols gave intermediate results.

These findings indicate that extensive optimization of protocols for recovering DNA from spent cartridge casings can result in far more STR data compared to earlier efforts. Given the very high probative value that spent cartridge casings may hold, and the frequency at which they are recovered from shooting scenes, these findings will be of considerable use to crime laboratories and the criminal justice system.

Cartridge Casings, Touch DNA, DNA Extraction

A113 Optimizing Transportation and DNA Extraction Methods From Fingernail Evidence

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After attending this presentation, attendees will gain insight into better methods of packaging evidence for transportation from the crime scene to the forensic laboratory. They will also better understand processing fingernail evidence, particularly enhancing the recovery of exogenous (foreign) DNA while minimizing recovery of endogenous (fingernail) DNA.

This presentation will impact the forensic science community as practitioners will learn more about the process of transporting and extracting DNA from fingernail evidence. They will be introduced to methods for packaging fingernail evidence to minimize loss of exogenous material, so DNA degradation does not occur. They will also learn improved methods to recover exogenous DNA while reducing the amount of endogenous DNA from fingernails, which may reduce the level of mixtures in STR profiles.

During a physical assault, biological material can be transferred between an assailant and victim, and it is clearly

important to properly collect, transport, and analyze such evidence. When victims defend themselves with their hands, biological material from the assailant may accumulate underneath their fingernails; however, there might only be trace amounts of DNA present from the attacker. Thus, it is important to recover and retain as much of that exogenous material as possible. Research on fingernail evidence is limited, and although some has been conducted, there is no optimized method regarding fingernail evidence processing.¹ Therefore, it is unknown which type of packaging best prevents the loss and degradation of DNA, and what procedures are best for recovering the maximum amount of exogenous DNA while limiting the amount of endogenous (nail) DNA.

The current study involved deposition of 1 μ L of male blood on clipped female fingernails, which was dried overnight. This allowed easy determination of exogenous DNA loss through the quantification of male DNA. Blood deposition also controlled the amount of exogenous DNA on the fingernails, which actual scratchings do not allow. In subsequent studies, scratchings were examined in order to better mimic fingernail evidence from an assault.

Fingernails harboring blood were pre-soaked in one of the following: phosphate buffered saline; Tris/EDTA, digestion buffer (Tris, EDTA, SDS); tissue lysis buffer from a Qiagen[®] QIAamp[®] DNA Investigator Kit; or water. Soak time (5, 10, 20, 30, 60, and 120 minutes) and agitation during the soaking process were also investigated. During DNA extraction, agitation and various digestion times (30, 60, 120 minutes, and overnight) were compared. Total DNA (endogenous + exogenous) and exogenous (male) DNA were quantified.

To examine DNA loss during transportation, blood was again deposited on fingernails and dried. Nails were placed into coin envelopes, bindle paper, or microfuge tubes and transported for five days. The coin envelopes and bindle papers were swabbed to recover any loose exogenous material. Nails transported in the microfuge tubes were processed within that same tube. DNA was subsequently extracted and quantified. The degradation of DNA on nails stored in microfuge tubes over time was also examined, storing them for two days, one week, two weeks, one month, three months, and six months. Again, the samples were extracted and male DNA quantified.

Finally, scratchings were performed as follows: female volunteer scratched a male volunteer using her three middle fingers three times with two pounds of force (measured using a kitchen scale). The female volunteer's fingernails were clipped then placed into the different package types and transported for five days. The coin envelopes and bindle papers were again swabbed, and all samples were once again extracted and quantified. Autosomal and Y-STR analyses were also performed, in order to examine the utility of each procedure on this type of evidence.

Pre-soaking nails in digestion buffer (containing SDS) removed the most exogenous cells, although more endogenous DNA was released as well. Agitation during pre-soaking helped in exogenous DNA recovery; however, agitation during digestion was disadvantageous, in that large quantities of endogenous nail DNA were also released. During transportation, exogenous DNA was lost from nails, which was not recoverable through swabbing the packaging. In this regard, transporting nails in a container such as a microfuge tube, where DNA both adhering to the nails and any lost component can be co-recovered, is advantageous, although considerable attention needs to be paid to DNA degradation. Scratchings result in far less exogenous DNA recovery; however, STR profiles can be produced. In these samples, mixtures are often apparent, which in these tests could be circumvented using a Y-STR assay.

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DNA Evidence, Fingernail Evidence, Evidence Packaging

A114 A Touch-ing Retrospective: A Historical Review of Six Years of Touch DNA Case Results

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After attending this presentation, attendees will benefit from scientific observations on the DNA yield, genetic yield, and information yield for a variety of different sources of touch DNA. Due to increases in the sensitivity of the amplification kits currently utilized by the forensic biology field, samples have expanded past traditional stain level sources. Submission of low level, or touch, DNA samples is increasing across the forensic community but, to date, there is a lack of comprehensive data on the best sources of useable touch DNA.

This presentation will impact the forensic science community by providing guidance for casework management of touch DNA samples so that laboratory resources are focused on samples which are more likely to provide higher quality, useable data. Because backlogs are a constant companion to the forensic science laboratory, efficiency is essential in proper casework management to maximize limited resources. Moreover, the information derived from this study can provide serviceable information for courtroom testimony.

Touch DNA has become a very useful source of evidence. In many situations, it has provided physical evidence in cases where there is a lack of a traditional, stain-level source of DNA. As a result, submissions of challenging, low-level samples have significantly increased in laboratories nationwide. This advent of touch sample submissions has enlarged backlogs due to burgeoning sample numbers as well as increased time for analysis completion. Guidance for appropriate casework management of the various sources of touch DNA can aid in reducing inflated backlogs created by the new touch DNA evidence stream. Additionally, the success and failure of touch DNA samples is often a point of contention during testimony. A scientific analysis of results can provide useful supporting information when testifying on the results obtained for touch DNA samples.

Data indicating the DNA yield, genetic yield, and information yield were recorded for each touch DNA item processed by the Allegheny County Office of the Medical Examiner Forensic Laboratory from 2008 to 2013. DNA yield, the quantity of DNA obtained from a sample, was measured as the quantity of DNA obtained from each sample using real-time Polymerase Chain Reaction (PCR). Genetic yield is the measurable quality of the profile obtained for the low level sample. Genetic yield was determined by the peak quality and number of alleles obtained from each sample through analysis with capillary electrophoresis. Finally, the information yield, which is the measurement of the utility of the profile, was measured by the completeness of the profile, resolvability of the mixture, if present, and the statistics obtained, if references were available. This information was grouped according to sample type, in an effort to negate the influence of varying collection methods between sample types. Sample types included firearms, magazines, baseball caps, masks, car door handles,

doorknobs, shirts, and gloves. The data was then analyzed to determine trends associated with the various sample types.

As expected, the best sources of touch DNA were firearms and clothing articles. Most likely due to the number of textured areas and the force required to operate, firearms provided not only the highest levels of DNA quantity, but also afforded the most usable information in terms of resolvable and simple mixtures. Firearms samples also afforded a higher incidence of Combined DNA Index System- (CODIS) suitable profiles. Shirts and hats, likely due to prolonged wearing, provided results similar to those from firearms. Less successful results were obtained from weapons (other than firearms), tools, gloves, and masks. The DNA yield for these types of items was significantly lower than anticipated for types of items subjected to prolonged or forceful handling. Of note, information yield is appreciably increased in cases with known references. The sample types with low levels of DNA yield, genetic yield, and, ultimately, information yield were: door knobs; car door handles; paper; magazines; and bags. The majority of these sample types resulted in partial DNA profiles or no amplified product at all.

Data utilized in this study was obtained, in part, from samples processed with instrumentation and chemistries purchased with the support of the National Institute of Justice 2008 DNA Unit Efficiency Grant (2008-DN-BX-K191), the 2009 DNA Backlog Reduction Grant (2009-DN-BX-K125), and the 2010 DNA Backlog Reduction Grant (2010-DN-BX-K065). The opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect those of the Department of Justice.

Touch DNA, Casework Management, Testimony

A115 Enzymatic Means to Rehabilitate Low-Copy Number DNA

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After attending this presentation, attendees will gain an understanding of the difficulties associated with processing Low-Copy Number (LCN) DNA, the impact these difficulties have on profile determination, and how these problems may be alleviated enzymatically.

This presentation will impact the forensic science community by providing a method to generate a full forensic DNA profile from minimal amounts of biological sample using an enzyme known as Duplex-Specific Nuclease (DSN).

It has become a routine practice to utilize DNA analysis in the investigation of violent crimes and sexual assaults. The success and acceptance of this methodology has led law enforcement agencies to turn to DNA analysis for cold cases, kidnappings, property crimes, and other crimes where additional types of evidence are insufficient. Most commonly, evidence from these cases are skin cells left on some sort of contact item that contain very little DNA. The analysis of LCN DNA is perhaps the greatest challenge to forensic DNA analysis, because the lack of sufficient DNA usually results in profiles that are difficult or impossible to interpret. Ultimately, this could lead to the false inclusion or exclusion of a suspect.

Currently, the most common procedure performed to analyze low-copy number DNA is increasing the number of Polymerase Chain Reaction (PCR) amplification cycles. Several experiments have been performed on DNA samples containing less than 100pg of DNA using 34 amplification cycles instead of the standard 28. Despite this methodology's potential to generate a more complete profile, it is unlikely that a protocol for

an increase in PCR cycle number will be routinely utilized in the forensic community. *Taq* DNA polymerase begins to degrade at increased cycle numbers, making it inefficient for DNA replication¹ Additionally, current LCN protocols state that an allele can only be reported if it is observed in replicate samples. As stated previously, case samples containing LCN DNA often contain small starting amounts of DNA, potentially making multiple runs unlikely.

The DSN enzyme has been successfully used in research settings to digest double-stranded DNA in order to normalize samples containing unequal representations of template.² This enzyme, found in the Kamchatka crab, is highly thermostable and is active over a wide pH range.³ The addition of DSN to a DNA sample takes advantage of the reassociation of denatured DNA in that sequences that are more abundant in a sample find their complementary strand at a much faster rate than those with fewer copies.⁴ DSN can be used to cleave DNA strands that have reannealed, thereby reducing the amount of over-represented sequences. This methodology allows for the lower concentration sequences to become equalized within the sample in a manner that is similar to the normalization of sequencing libraries performed before next-generation sequencing.⁴

The ability to generate a full profile from minimal amounts of sample could prove to be an invaluable resource when attempting to provide information during a forensic investigation. Therefore, the application of DSN to normalize low-copy number DNA concentrations in biological samples could be useful when applied to forensic DNA profiling. In conclusion, it is hypothesized that the addition of DSN will alleviate the preferential amplification of smaller alleles, thereby allowing equal representation of all alleles analyzed and a more complete forensic profile to be generated.

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Low-Copy Number, DNA, Duplex-Specific Nuclease

A116 Examination of Proposed Manufacturing Standards Using Low Template DNA

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After attending this presentation, attendees will understand sensitivity differences between quantitative Polymerase Chain Reaction (qPCR) and Short Tandem Repeat (STR) analysis. In addition, attendees will understand why both analysis methods are suitable in manufacturing process for the purpose of certifying product as Forensic Grade.

This presentation will impact the forensic science community by allowing the community to understand testing constraints in manufacturing processes and why multiple analysis methods are needed for Forensic Grade certification.

Forensic DNA laboratories rely on reagent and plastics manufacturers to supply high-quality products with minimal interference from contaminating DNA. With the increasing sensitivity of STR amplification systems, levels of DNA that were previously undetected may now generate partial profiles. To address the concern of laboratories worldwide regarding the potential of low-level DNA contamination in consumables, accrediting bodies in the United Kingdom and Australia proposed guidelines PAS377 and ISO 18385, respectively, for minimizing the risk of human DNA contamination events during the manufacturing process.

The guidelines also propose the acceptable limits for "contaminating DNA" as well as the methods for detecting the potential contaminant. The United Kingdom guideline recommends using only STR testing, while the Australian guideline allows both STR and qPCR as suitable methods. This presentation compares the sensitivity of qPCR to STR analysis and discusses the suitability of each method in the manufacturing process for the purpose of certifying a product as Forensic Grade.

To determine the sensitivity of STR analysis, the sensitivity of its two major components was analyzed: the Capillary Electrophoresis (CE) instrument and the STR reagents. To determine the sensitivity of the CE instrument, a high amount of DNA (500pg) was amplified to eliminate the stochastic effect of amplification of low template DNA amount. This ensured that any dropouts at low input amount are due to CE limitation and not PCR variability. Instrument sensitivity was tested using default and enhanced conditions as recommended by the United Kingdom guideline: longer injection and lower peak calling threshold. Under enhanced conditions, the Limit Of Detection (LOD) for the CE instrument is 0.5pg.

The United Kingdom guideline also allows for replicate analysis as performed in Low-Copy Number (LCN) analysis. Using DNA input titration followed by analysis under LCN analysis (i.e., repeat injection followed by consensus allele calling), sensitivity down to 1pg can be achieved. With input DNA of 5-10pg, which is approximately equivalent to the amount of DNA in one cell, 30-60% of the alleles were called. This would not meet the criteria suggested by the proposed guidelines for being Forensic Grade.

In contrast to STR analysis, qPCR analysis is sensitive down to 0.25pg of input DNA. In addition to increased sensitivity, qPCR analysis is more suitable for testing a higher number of samples in that it is more cost effective and data interpretation is simpler. Testing a large sample number is necessary for increased statistical confidence in a destructive test where a representative sample from each batch is tested and destroyed.

Therefore, it is proposed that qPCR analysis be used for testing plastic consumables. For STR reagents, using STR analysis is proposed as it will simultaneously test all components of the kit for the presence of contaminating DNA. While this presentation discusses the LOD for the test methods, the limit that is acceptable to forensic laboratories still needs to be determined.

STR, Manufacturing Standards, DNA Contamination

A117 Developing STR Profiles From Fired and Unfired Brass and Nickel-Plated Cartridge Cases

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After attending this presentation, attendees will be aware of the likelihood of obtaining Short Tandem Repeat (STR) DNA results from nickel-plated and brass cartridge cases, both unfired and fired.

This presentation will impact the forensic science community by examining the ability forensic biologists have to recover useful DNA profiles from fired and unfired cartridge cases depending on metal composition by analyzing and comparing both nickel-plated and brass cartridge cases.

DNA is transferred to ammunition when it is being loaded into a firearm by shed skin cells found in the oil and sweat residues of fingerprints; however, obtaining complete DNA profiles from cartridge cases can be difficult because the DNA is present in small quantities or has been degraded. It has been surmised by members of the forensic science community that the amount of DNA that can be recovered depends on the type of cartridge case used. It is commonly held that there is less of a chance of obtaining useful DNA profiles from brass cartridge cases than from nickel-plated or aluminum cartridge cases; however, this is not well documented in published literature. There are many theories as to why it could be more challenging to recover DNA from brass cartridge cases in comparison to nickel-plated or aluminum cartridge cases; however, these theories have either been disproved or not thoroughly tested. This research used a sample size of 200 cartridge cases to determine the frequency with which nuclear DNA could be recovered from both fired and unfired brass and nickel-plated cartridges.

Of the 200 cartridge cases used in this study, 100 were brass and 100 were nickel-plated. All cartridge cases were the same caliber, 45mm. The cartridge cases were handled only long enough to load into a 10-round magazine. Half of these cartridges were fired and half were unloaded from the magazine manually. The fired and unfired cartridges were swabbed for DNA using a double swab technique. All samples were extracted using a Qiagen® EZ1® automated large volume protocol, quantitated using real-time quantitative PCR, amplified using the Promega® PowerPlex® 16 HS Amplification Kit, and analyzed using an Applied Biosystems® 310 Genetic Analyzer.

No statistically significant difference in quantity of DNA recovered from cartridge cases as a function of metal type was observed. However, DNA was more frequently recovered from brass cartridge cases than from nickel-plated. During quantitation, detectable levels of DNA were recovered 36% of the time from brass cartridges and 12% of the time from nickel-plated cartridges. The unfired brass cartridges yielded partial DNA profiles 36% of the time while unfired nickel-plated cartridges yielded partial DNA

profiles only 12% of the time. No profile could be obtained from the remaining brass and nickel-plated cartridges. This study demonstrates the difficulty of producing DNA profiles from cartridge cases regardless of metal type. Work to optimize DNA recovery based on collection technique is currently in progress.

Forensic Science, STR Analysis, Cartridge Cases

A118 Rapid PCR Using a Microwave Oven: From Sample to Profile in Less Than One Hour

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After attending this presentation, attendees will learn about a less-than-two-minute extraction method followed by a rapid Polymerase Chain Reaction (PCR) approach that shortens the time to generate a Short Tandem Repeat (STR) profile from a buccal swab (reference sample) to less than an hour, using Chelex® 100, a microwave oven, a Philisa® Thermal Cycler, and a 3130 Genetic Analyzer.

This presentation will impact the forensic science community by providing an alternative to microfluidic devices and automated systems. The approach uses conventional technology, for the most part already available in forensic DNA laboratories, to generate profiles with speed, accuracy, and reproducibility at a lower cost.

The Philisa® Thermal Cycler has a ramping speed of 12-15°C/sec, with reaction volumes between 10-50µL, and is significantly less expensive than the recently developed sample to profile automated systems. It uses specially designed high-quality polypropylene tubes with a thin wall for efficient and uniform heat transfer. These tubes, in combination with the fast ramping speed of the thermal cycler, allow for significant reduction in amplification time; however, in order to effectively take advantage of the faster amplification time, a comparably fast DNA polymerase is necessary for the reaction.

Chelex® extraction is a conventionally used, inexpensive, and efficient method to extract DNA that requires heat (a boiling step). In this approach, extraction from a buccal swab was performed by heating it in a microwave oven. Microwave ovens use electromagnetic radiation to heat food. Since microwaves are sinusoidal waves having peaks and troughs, the intensity of heat varies along the wave. Hence, microwave ovens are provided with a rotating aid that rotates the food, allowing uniform heating. In the absence of the rotating aid, the intensity of heat is concentrated in specific locations inside the microwave oven. In this study, these specific locations were identified using shredded cheese.

Shredded cheese was added uniformly onto wax paper and placed in a commercial Sharp® Carousel® microwave oven without the rotating aid. The melting pattern of the cheese was noted at 30, 90, and 150 seconds. To check for reproducibility, the procedure was repeated while maintaining the thickness of the cheese layer after allowing the plate to cool. The cheese started melting first in the right-hand corner of the oven in all three trials. This location was marked and referred to as the "hotspot".

All extractions were performed by placing the buccal swabs in 500µL of 5% Chelex® 100 solution in screw-cap tubes. The tubes were vortexed for 10 seconds and placed on the hotspot of the oven without the rotating aid. Extraction was then performed by starting the timer on the oven and selecting maximum power (1000W). Different lengths of time were evaluated (1, 1½, 2, 2½, and 3 minutes).

The polymerase selected for this study is SpeedSTAR™. In a final volume of 11.25µL, 5µL of the Premix Ex Taq™ (2X) (Perfect real time) PCR reaction mix was combined with 0.5µL of the SpeedSTAR™ enzyme (used as a booster), 2µL of AmpF/STR® Identifier® Primer Mix, 0.75µL of BSA (625ng/µL) and 3µL of diluted extract (1:2.5 dilution). Amplification was performed in a two-step protocol consisting of 28 cycles of 5 seconds at 95°C and 20 seconds at 58°C, with an initial denaturation at 95°C for 30 seconds and a final hold at 15°C for 60 seconds.

Within the data set generated by analyzing samples with the most recently optimized parameters, the observed full locus drop-out rate was 5%. The overall heterozygous peak balance ranged between 0.47-1.00, with an average of 0.8. The signal intensity ranged between 40-4000RFU, with an average of 390RFU. Artifacts such as stutter and incomplete adenylation were observed.

The integration of Chelex® extraction in a microwave oven and PCR amplification in the Philisa® Thermal Cycler with SpeedSTAR™ polymerase has allowed generation of a DNA profile with a sample to profile time of less than one hour. This approach provides speed while maintaining reproducibility and accuracy, all while using conventional methods of analysis that can be implemented in any laboratory setting. The disadvantage of this system is that it is not specifically designed for non-experts.

Rapid PCR, Microwave Extraction, STR analysis

A119 Incorporation of a 24-Locus STR Assay Into a Fully Integrated Rapid DNA Analysis System

James W. Schumm, PhD, 13315 Palmers Creek Terrace, Lakewood Ranch, FL 34202-5005*

After attending this presentation, attendees will learn that a 24-locus Short Tandem Repeat (STR) multiplex system has been adapted to rapid DNA analysis and is compatible with use in fully automated instrument systems.

This presentation will impact the forensic science community by discussing new capabilities for rapid DNA analyses based on expanded STR sets that will enhance DNA profiling outside the laboratory in office and field-forward settings.

The Department of Homeland Security has long recognized the need to expand the number of STR loci used in its immigration services, refugee-asylee evaluations, and other efforts, and the value of moving these operations outside the laboratory to distributed office environments and field-forward locations. With the Federal Bureau of Investigation's (FBI's) proposed expansion of the core STR set to be used in the Combined DNA Index System (CODIS) national database and their programs to introduce rapid DNA analysis into forensic laboratories and police stations, the need for an automated easy-to-use rapid DNA analysis system has become increasingly clear.

A rapid DNA analysis system consisting of a ruggedized instrument and a single-use BioChipSet™ Cassette (BCSC) has been developed.¹ The cassette accepts five buccal samples, automatically performs DNA purification, amplification, electrophoretic separation and detection of amplified products, and assigns correct alleles (based on the Promega® PowerPlex® 16 assay) to create DNA profiles ready for additional evaluation or upload to the CODIS database.² The reagents are stable for at least six months at 22°C, and the instrument has been designed and tested to Military Standard 810F for shock and vibration. The results of performance assessments indicate that the integrated system generates concordant short tandem repeat profiles.³

This presentation describes the adaptation of the Promega®

PowerPlex® Fusion System to rapid DNA analysis. Maintaining the same primer sequences as the National DNA Index System (NDIS)-approved product enhances compatibility with previously completed population database studies and concordance evaluations. To adapt to rapid thermal cycling conditions, modifications were made to primer concentrations to generate a balanced 24-locus multiplex in a 19.5-minute amplification.

In addition to achieving balance, it was important to eliminate Polymerase Chain Reaction (PCR) artifacts. The sources of such artifacts were identified by serial removal of primer pairs, making it possible to lower specific primer pair concentrations to diminish or eliminate the offending artifact fragments without significant loss of signal of the correct allele fragments. It was not necessary to change the fusion primer sequences to remove artifacts in the rapid amplification assay.

A critical step in adapting successful rapid amplification protocols to full automation is the lyophilization of the PCR mix for incorporation and storage as a stable component of the BCSC. Data demonstrating stability of the lyophilized reaction mix will be presented. Finally, automated interpretation of the 24-locus profiles required adaptation of the expert system to pre-programmed analysis of the results using the 24-locus assay. This modification to the expert system will be discussed.

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1. GE Healthcare Life Sciences DNAscan Rapid DNA Analysis System, <http://www.gelifesciences.com/webapp/wcs/stores/servlet/productById/en/GELifeSciences/29022852>
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3. Tan E, Turingan RS, Hogan C, Vasantgadkar S, Palombo L, Schumm JW, and Selden RF. Fully Integrated, Fully Automated Generation of Short Tandem Repeat Profiles. *Investigative Genetics* 2013, Vol 4, No. 16, doi: 10.1186/2041-2223-4-16.

Rapid DNA, Microfluidics, STR Multiplex

A120 Rapid DNA Analysis System for STR Typing of Low DNA Content Samples

Eugene Tan, PhD, NetBio, 830 Winter Street, Waltham, MA 02451*

After attending this presentation, attendees will learn: (1) the differences between automated microfluidic purification of reference samples and low content DNA samples; (2) an approach to the incorporation of a highly efficient low content DNA purification module into a fully integrated Rapid DNA Analysis system that enables forensic sample analysis to be performed rapidly in field forward settings; and, (3) the dynamic range and limit of detection of the fully integrated low DNA content system.

This presentation will impact the forensic science community by discussing the potential for fully integrated, samples-in to results-out Short Tandem Repeat (STR) reference sample analysis systems to be used in a wide range of out-of-laboratory settings, including police stations, borders and ports, military checkpoints, and the battlefield. The expansion of sample types amenable to Rapid DNA Analysis to include those with low DNA content has potential utility in casework analysis in general and property crime analysis in particular.

NetBio has previously developed a fully automated Rapid DNA Analysis System that produces STR profiles from buccal and blood samples for human identification in 84 minutes with no manual processing steps or manipulations.¹⁻³ To expand the applications of the system, the integrated microfluidic biochip for reference sample processing (core BioChipSet Cassette (BCSC)) has been modified to process low DNA content samples. Buccal samples

contain >1000-fold excess DNA than required for amplification and, as such, the purification module in the buccal system discards 99% of the input sample DNA. Low DNA content samples (sometimes referred to as touch samples) include those that result from objects coming in contact with an individual, including cigarette butts, drinking glasses, bottles, cans, firearms, clothing, and fingerprints. The variety of these sample types is reflected in the wide range of their DNA content, generally ranging from less than 0.1 to 100ng. In contrast to reference samples, touch samples require a modified purification system that retains essentially all the input sample DNA. The aim of this presentation is to present the design and function of the touch sample BCSC.

With the goal of maximizing the DNA content for downstream processing, several purification steps were optimized and high-efficiency DNA-capture features were incorporated. Several swab types were assessed to identify the most effective choice for cell capture while retaining minimum lysate. Cell lysis, wash, and elution steps of the DNA purification protocol were optimized and several DNA purification filter types and sizes were evaluated for improved binding capacity and elution efficiency. In addition, a microfluidic ultrafiltration module was developed to concentrate the eluted DNA into a small volume so almost all of it could advance to PCR.

Optimization of purification protocols, filter type, and filter size resulted in DNA recovery of >90% with 50ng input DNA and near 100% with 1ng input DNA. The ultrafiltration module was then incorporated into the BCSC. The results of these modifications have achieved near 100% recovery of input sample DNA. The touch sample BCSC has retained the same form factor as the buccal BCSC and is fully compatible with the fully integrated instrument.

Data demonstrating the functionality of the low DNA content purification module and the fully integrated, fully automated touch sample BCSC will be presented. Issues related to rapid DNA analysis of touch samples, including the role of quantitation and minimization of contamination, will be discussed.

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Rapid DNA, Microfluidics, Low DNA Content Sample

A121 Fully Integrated Microfluidic Cartridge for Forensic STR Analysis of Reference Swabs

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After attending this presentation, attendees will see how it is possible to adapt the conventional entire process of Short Tandem Repeat (STR) DNA typing into a small microfluidic chip and obtain results with commercial kits. The quality of these results will be shown, taking into account forensic laboratories criteria.

This presentation will impact the forensic science community by demonstrating that an unskilled user can develop DNA profiles from reference swabs using a fully integrated device with a single consumable and getting results comparable to conventional processes obtained by a skilled operator in a laboratory.

DNA analysis is currently considered to be the gold standard for Human Identification (HID). Currently, the goal is to reduce time-to-result, cost, and analyst time consumed in processing reference samples. This would free the analyst for more complex casework and reduce the number of samples contributing to backlogs. The possibility of having the analysis performed directly by a police officer or inexperienced operator directly in an office may reduce the time from collection to entry in a database. Additionally, an integrated platform with a single consumable requires less logistic support for laboratory or office environments. It is for this reason that an instrument capable of generating DNA results from buccal swabs in less than two hours has been developed, exploiting microfluidics in a single, disposable cartridge that facilitates fully-automated analysis of four samples.¹ The cartridge interfaces with a single instrument capable of performing all steps necessary for generating STR profiles from four buccal swabs simultaneously, with no human interaction with either the instrument or the software during the analysis.

The goal of this study was to evaluate the quality of the subassays and to assess the performance of the fully integrated DNA instrument run by an inexperienced operator.

A set of 50 sample-to-result analyses were performed and the performance of the subassay processes for these runs was evaluated. The first step evaluated was the DNA extraction performed by the platform using the ZyGEM[®] EA1 enzyme for liquid phase DNA preparation. Results showed that the DNA yield obtained was sufficient to allow good DNA amplification for all samples. In the 50 samples run, infrared-mediated Polymerase Chain Reaction (PCR) amplification resulted in 94% full profiles (18/18 loci) and 6% partial profiles (17/18 loci) when the product was analyzed on an Applied Biosystems[®] 310 Genetic Analyzer. Finally, the efficiency of microchip electrophoresis was demonstrated with a series of allelic ladders (five cartridges) run over five weeks. Precision of the separation was always within the +0.5/-0.5bp range.

In the last part of this study, results showed that the platform could successfully integrate all of the processes using a single, simple cartridge. The presented results of the entire integration show data calculated from a set of more than 50 samples obtained with Promega[®] PowerPlex[®] 18D reagents generated by an unskilled operator. The results showed that allele calling from integrated analyses had 100% concordance with the conventionally generated profiles when peaks were observed. Typical forensic quality metrics for these analyses will be presented. The results showed stutter ratios less than 20% and heterozygote peak balance greater than 60% for 94% of the samples.

While work continues to improve the data quality and robustness of the platform, results indicate that full integration of the laboratory processes into a single, disposable HID microdevice is

possible. This allows for automated sample-to-answer capabilities when processed by an inexperienced operator and begins to deliver on the time, cost, and analysis time benefits promised by microfluidics.

Reference:

1. Root, B *et al.* A Multichannel Microdevice for PCR Amplification and Electrophoretic Separation of DNA. Proceedings of 15th International Conference on Miniaturized Systems for Chemistry and Life Sciences (μ TAS), 2011, Seattle, WA.

Rapid DNA, Microfluidic Cartridge, STR Analysis

A122 Glycol Ethers: A New Category of Oxygenated Solvents Encountered in Fire Debris Samples

Troy J. Ernst, MS, Michigan State Police, Grand Rapids Forensic Lab, 720 Fuller Avenue, NE, Grand Rapids, MI 49503; and Kevin D. Streeter, BS, Michigan State Police, 720 Fuller Avenue, NE, Grand Rapids, MI 49503*

After attending this presentation, attendees will be informed of a category of ignitable oxygenated solvents — glycol ethers — that may be encountered when processing fire debris using a passive headspace adsorption method with parameters typically employed in recovering and analyzing ignitable liquid residues. They will be provided with data from reference samples, data from consumer products containing glycol ethers, and casework data from fire debris containing glycol ethers. With this knowledge, fire debris analysts will be able to gather and analyze reference samples containing glycol ethers. This will allow them to identify and report the presence of glycol ethers in the fire debris so that fire investigators can properly assess the observations of the fire scene.

This presentation will impact the forensic science community by educating fire debris analysts of potentially ignitable products that have been overlooked when encountered in fire debris samples.

In early 2010, information was received regarding the possible involvement of scented oils containing glycol ethers in intentionally set and accidental fires. Based on comparisons of mass spectral library search results and information from Material Safety Data Sheets from scented oils, it was discovered that a case that had been recently analyzed in the laboratory may have contained these compounds. According to the American Society for Testing and Materials (ASTM) International ignitable liquid classification scheme (E1618-11), identification of an oxygenated solvent requires that all major oxygenated compounds be identified by gas chromatographic retention times and mass spectral characteristics.¹ Without a reference sample containing these compounds, a comparison of retention times cannot be performed; therefore, identification of that oxygenated solvent is not possible if one is to follow the standard. If an ignitable liquid is present, but the threshold for its identification is not met, the fire investigator does not receive information that may be probative to the investigation.

Consumer products containing glycol ethers — mostly plug-in room fresheners and vehicle deodorizers — were purchased for analysis. Additionally, reference standards of several glycol ethers were obtained from The Dow Chemical Company.² The consumer products and the glycol ether reference standards were subjected to flame tests and analyzed by Gas Chromatography/Mass Spectrometry (GC/MS). Some of the glycol ether reference standards showed agreement in retention times and mass spectra to the consumer products and to the case samples.

In the last three years, several case samples containing

oxygenated solvents consistent with glycol ether reference standards have been encountered. Laboratory reports have been issued that identified glycol ethers in fire debris samples. Identification of these compounds may provide fire investigators with valuable information regarding crime scene observations or fire causation.

References:

1. ASTM International, 100 Barr Harbor Drive, West Conshohocken, PA 19428-2959
2. The Dow Chemical Company, 2030 Willard H. Dow Center, Midland, MI 48674

Fire Debris, Glycol Ethers, Oxygenated Solvents

A123 Infrared Photographic Analysis of Blood Under a Variety of Paint Pigments With an Analysis of the Pigments Via Raman Spectroscopy

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After attending this presentation, attendees will understand an alternate method to chemiluminescence in the detection of blood underneath paint. It has already been established that products such as Bluestar® can be used to reveal hidden blood, but the technology does have difficulties associated with it that this photography technique does not.

This presentation will impact the forensic science community by: (1) expanding knowledge of the mechanics of Infrared (IR) photography and its applications toward forensic science; (2) explaining the technique and further applications of IR photography and how its analysis of blood spatter under paint can compare to the use of products such as Bluestar®; and, (3) educating the forensic science community as to how one can replicate the experiment under their own settings and requirements.

Detection of blood underneath paint is a unique and rarely encountered situation. In the case of an investigator's suspicion that blood has been painted over, destruction of the wall (and possible destruction of evidence) may occur. As such, a technique to detect the fluid underneath the paint layers without destruction of the material would be invaluable in saving time and effort, as well as cost. The price of an infrared-capable camera is not outrageous, even for the most cash-strapped investigation units. Previously purchased cameras can be modified to photograph in the infrared. Guides to do this can be found online, or alternatively one can send the camera to individuals who guarantee and perform the process for a modest fee. Training can be accomplished in a relatively short period, requiring only general photography skills. As with any other photography technique, experience can improve the work product.

Previous research into the topic yielded several different results. First, depending on the type and color of paint used, infrared photography could penetrate up to six layers of paint. Some colors, such as green, prevent transmission beyond layers. Red and purple were two colors that did not prevent penetration at all, providing a clear picture of applied bloodspatter underneath the paint. Blood was applied using a plastic stencil, with several copies of the spatter reproduced, one for each type of paint tested. Other research has indicated success with using this particular photography technique, but only three layers of paint were tested.

Infrared photography is a specialized technique that is not widely used (or needed) at crime scenes. Instances of blood being painted over at scenes are few and far between. More commonly,

the blood is simply cleaned up. There are methods to reveal blood that has been cleaned up, including the use of Bluestar® and luminol. This particular photography technique relies on a significant volume of blood to be present. Previous research into the topic showed that infrared photography will not pick up blood that is heavily diluted or cleaned up, but this was not investigated for this project. Small blood spots can be missed during a cleaning, time may be limited, or lack of cleaning supplies may prevent a thorough cleaning. It is then that an infrared photographic technique can be employed.

Research performed investigates the way infrared light interacts with the different paint pigments purchased or provided by Sherwin Williams®. The paints/pigments were analyzed using Raman spectroscopy, in order to compare them to Christopher Palenik's master sheet. A correlation is planned to be drawn between penetration and reflectance/absorbance of the pigments detected under Raman spectroscopy.

Photography, Pigment, Blood

A124 Determination of Pesticide Residues in Marijuana by QuEChERS and LC/MS/MS

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After attending this presentation, attendees will learn about the QuEChERS extraction and analysis of pesticides and their concentrations in samples of street-level marijuana and the relationship with cannabinoid levels in this material using Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS).

This presentation will impact the forensic science community by offering analysts operating in forensic facilities information regarding the QuEChERS extraction and analysis of popular cannabinoids encountered in drugs and by driving casework using solid phase extraction and LC/MS/MS.

Introduction: Marijuana is one of the most commonly used illicit drugs in the world. Although the concentrations of active ingredient (THC) may be determined by laboratories, few studies have looked at possible organic contaminants such as pesticides in seized marijuana samples. By developing a robust and efficient method using QuEChERS (Quick, Easy, Cheap, Effective, Rugged, and Safe) and LC/MS/MS techniques, the identities and concentrations of 23 pesticides, including methamidophos, carbendazim, dicofol, acetachlor, thiabendazole, DIMP, tebutiuron, simazine, carbaryl, atrazine, DEET, pyrimethanil, malathion, bifenthrin, tebuconazole, cyprodinil, diazinone, zoxamide, pyrazophos, profenofos, chlorpyrifos, abamectin, and bifenthrin were determined.

Objective: This project was developed to determine the identities and concentrations of pesticides in street-level marijuana using a robust and efficient methodology. In this study, QuEChERS method was employed in conjunction with LC/MS/MS for pesticide analysis.

Method: To each calibrator and test sample (2g) was added 10mL of Deionized (DI) water. Samples were mechanically shaken for 60 minutes, after which the internal standard (triphenyl phosphate (1µg)) was added. To the samples were added 10mL of acetonitrile, and 4g of magnesium sulfate (MgSO₄)/ 2g of sodium chloride. The mixture was shaken for one minute. These samples were centrifuged for five minutes at 5,000rpm. A 2mL aliquot of the upper layer was transferred to a dispersive Solid Phase Extraction (dSPE) tube containing 150mg MgSO₄, 50mg Primary Secondary Amine (PSA), and 50mg ChloroFiltr®. The tubes were shaken for 30 seconds, and then centrifuged for five minutes at 10,000rpm.

A 300µL aliquot was transferred to an autosampler vial and mixed with 300µL of DI water and filtered before analyzing by LC/MS/MS. Matrix matched standards were made by spiking varying amounts (20-4,000ng) of target pesticides to the marijuana extracts (external calibration).

Details are presented for the liquid chromatography performed in gradient mode using: (1) DI water containing 0.3% ammonium formate and 0.1% formic acid; and, (2) methanol with 0.1% formic acid using a Thermo™ Accucore™ aQ LC column (100 x 2.1mm, 2.6µm) and tandem mass spectrometry with ESI+.

Results: This method was found to be linear from 10-2,000ng/g, ($r^2>0.995$) with a Limits of Detection/Quantification (LOD/LOQ) of 5ng/g and 10ng/g. Recoveries of the pesticides ranged from 72.5%-149.2%. The majority of pesticides lay within 70-120%. %CV for the analysis was less than 20%. This data was achieved by an external calibration procedure, where the analytes were measured against matrix extracted but externally fortified samples. This newly developed method was applied to ten real marijuana samples released to UCT.

Conclusion: Pesticide residues were detected in all of the marijuana samples tested. Methamidophos, carbendazim, acetachlor, DEET, and chlorpyrifos were among the most often detected pesticides in this study. Because there are currently no pesticide threshold values established for marijuana samples, the detected pesticide levels were compared to the European Union maximum residue levels for foodstuff (tea) samples (10ng/g to 5,000ng/g). Some pesticides were found that exceeded these levels, with a few at extremely high concentrations, such as 67,070ng/g carbendazim found in sample #8, and 75,137ng/g chlorpyrifos in sample #7. The findings indicated that pesticides were used for higher marijuana production, thus medical marijuana should be inspected for pesticide residues before the use by patients who suffer from HIV, AIDS, and cancers.

Pesticides, Quechers, Analysis

A125 Chemical Profiling of Trichloroisocyanuric Acid- (TCCA) Based Explosives for the Forensic Attribution of Precursor Materials

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After attending this presentation, attendees will gain an understanding of chlorinator-based explosives and how forensic trace signature profiles can be used to determine the source of the components used to make the explosive.

This research will impact the forensic science community by introducing a new method to analyze trace signatures of chlorinator-based explosives which may generate leads during an investigation. The analytical techniques presented here can potentially be applied to evidence derived from other types of homemade chemical explosives.

The use of homemade explosives poses a real and growing threat in the United States. Recently, explosives in which the primary oxidant is swimming pool chlorinator, typically Trichloroisocyanuric Acid (TCCA), and the accelerant is automotive brake fluid have come to the attention from the forensic community. The components are inexpensive and can be easily acquired from a variety of commercial outlets (i.e., automotive and hardware stores). Chlorinator-based explosives are simple to manufacture

and will self-ignite. Despite their potential to be used for illicit activities, few forensic signatures exist for analyzing residues from chlorinator bombs for source attribution. Trace chemical profiling is a useful strategy for analyzing chlorinator-based explosives. Previous research with high explosives such as Triacetone Triperoxide (TATP) has shown that precursor materials produced by different manufacturers have distinct chemical properties (metal species, volatile organic compounds) and can be used to identify the brand or commercial source of specific components from the residue.

The goal of this research was to analyze trace chemical signatures of different sources of swimming pool chlorinator tablets and automotive brake fluid. Two types of signatures were investigated, metal composition and volatile organic compounds. Five different brands of chlorinator tablets and three different brands of brake fluid were acquired from local hardware stores. The brake fluid was extracted in diethyl ether while the chlorine tablets were prepared in nitric acid for analysis. The metal concentration of the chlorinator tablets was analyzed with Inductively Coupled Plasma Mass Spectrometry (ICP/MS) and Scanning Electron Microscopy (SEM). Organic signatures from the brake fluid were profiled with Gas Chromatography Mass Spectrometry (GC/MS). Trace chemical profiles were also obtained for the post-blast explosive residues. The pre- and post-signature profiles were compared in order to determine if the metal and organic compounds remained after ignition. Principal Component Analysis was then performed to determine if different sources of pool chlorinator and automotive brake fluid could be discriminated based on their chemical profiles.

Results showed that different sources of brake fluid had unique GC/MS profiles. For example, MS analysis of the chromatogram peak at the retention time of 9.33min corresponded to octaethylene glycol and was only observed in one brand of brake fluid. Also, the chromatogram peak at Retention Time (RT) 14.44min, which indicated hexaethylene glycol dimethyl ether, was present only in another source of brake fluid. The relative abundances of some of the peaks varied significantly between the brake fluid sources as well. For example, the chromatogram peak at RT 12.36min showed a relative abundance of 38% for one brake fluid brand and 63% for the other two brands. In addition the chromatogram peak at RT 12.59min showed relative abundance values of 2%, 14%, and 31% across the three sources. These differences were also observed in the GC/MS profiles of the post-blast explosive residues. Many of the same retention times were present that corresponded to the same ethanol derivatives and glycol compounds. This indicates that some of the organic compounds in the brake fluid persisted after the explosive reaction. ICP/MS analysis also revealed that among different brands of TCCA pool chlorine there is significant variation in the concentration of certain metals such as copper, zinc, and magnesium. Thus, this research poses promising forensic trace signature profiles for both TCCA swimming pool chlorine and automotive brake fluid that are used in homemade chlorinator explosives.

Chemical Signatures, Attribution, TCCA

A126 Forensic Botany: Molecular Identification of Pinus Pollen Grains in Surface Soil

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After attending this presentation, attendees will understand how to identify single pollen grains in surface soils using a molecular approach.

This presentation will impact the forensic science community by providing a protocol for genotyping single pollen grains from surface soil. This will allow laboratories to use DNA typing of pollen grains as an alternative tool for discrimination of soil samples.

Soils are a valuable tool in forensic investigations as they contain organic and inorganic signatures relating to origin that can provide investigative intelligence and evidentiary value. Soil evidence has been extensively analyzed through testing of different properties such as color, particle size, organic, and inorganic content; however, it is not always possible to discriminate soil samples based on these properties alone, and therefore benefits may be had from alternative methods that provide additional information. In recent years, the analysis of soils using molecular tools has been the focus of several studies. These studies are primarily based on the analysis of the composition of bacterial communities present in soils using terminal Restriction Fragment Polymorphisms (t-RFLP) as a primary method. Although t-RFLP is a cost-effective technique and is capable of analyzing small samples, it neither has a high resolution power nor does it provide a strong statistical assessment for association. Based on the limited success of this approach, the use of a well-established method for DNA identification such as Short Tandem Repeats (STRs) should be considered.

Pollen identification is crucial for palynologists. Traditional palynological methods for the identification of pollen in surface soil can resolve at best to the generic level. By sequencing the Internal Transcribed Spacer (ITS) regions of nuclear ribosomal DNA, it is possible to identify *Chenopodiaceae* pollen grains from soil to the species level. Here, a molecular approach for the individual identification of *Pinus* pollen grains in surface soils was developed. Surface soil samples were collected from a region of southeast Texas. Fresh needles of 40 *Pinus echinata* were sampled for DNA genotyping, establishing a database of eight STRs. Individual pine pollen grains in a soil sample were separated from the soil and eight STRs of each pollen grain were amplified using a fluorescent multiplex approach. By comparing the amplified products to those in the *Pinus* database, it was possible to identify the pollen in the soil samples. This new technique provides a technical reference for molecular identification of surface soil pollen for other species. This work is necessary for further efforts to interpret the composition of surface soil pollen related to existent vegetation characteristics. It also has significant potential for enhancing the ability to identify pollen in criminal casework.

Short Tandem Repeats, Pollen, Soil

A127 A Survey on the Microscopical and Chemical Analysis of Synthetic Wig Fibers

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After attending this presentation, attendees will learn about the most discriminating features, both microscopical and chemical, of synthetic wigs. Synthetic wig fibers are manufactured to take on the appearance of natural hair for either cosmetic or costume use. Subsequently, the characteristics of wig fibers, especially as a collective group within a single wig, can vary greatly from those synthetic fibers collected from garments and household items.

This presentation will impact the forensic science community by providing the most discriminatory analytical sequence for synthetic wig fibers.

Consider a set of hairs that were recovered from the hairbrush of a victim. The analyst, however, discovers the hairs are in fact synthetic fibers that look like human hair. The analyst may wish to then determine the wig type, and ideally the manufacturer, to aid in the investigation. Eventually, the analyst may have to perform a comparison in order to determine if a seized wig made up of as many as ten different fiber types may have been the source of the few recovered fibers. Fortunately, wig evidence may be recovered as long fibers, usually about 15cm. This is an advantage because the quantity of evidence will be large enough to permit destructive testing. The goal of this study is to determine what characteristics of wigs are polymorph; and to develop the most discriminating analytical sequence for microscopic and chemical examinations of wig fibers for the detection of the most variable features.

For this study, 49 new or slightly used synthetic wigs were donated by local retailers: 31 brown; 5 black; and, 13 blonde. Fiber samples from each wig were first examined visually and microscopically. The physical properties of the fibers documented include: longitudinal appearance; surface topography; color, thickness; cross-sectional shape, and, density of the delustrant particles.

In a previous phase of this study, there was found to be important overlap in the fibers' thickness and cross-sectional surface area measurements between wigs; however, it was observed that the cross-sectional shape is a variable feature that could increase the discriminating power of the analytical sequence. The Joliff method for creating cross sections utilized in this study showed that as many as six different cross-sectional shapes can be observed within a single wig. It was also observed that as many as eight different colored fibers can be combined within a wig. It is the selected combination of shapes and colors within a wig that adds to its selective profile. Thin-Layer Chromatography (TLC) was used to further study the dye content of the colored fibers. The dye extraction resulted in information about the dye type according to its application mode: acid; basic; or, disperse. The consecutive dye elution then provided information about the variation of dyes between different wigs.

Indications of the fiber types were achieved through the initial fiber assessment by means of their optical properties. These include their degree of birefringence and sign of elongation. According to the literature, nylon, polyester, polypropylene, acrylic, and modacrylic fibers are the most commonly used fibers, with modacrylic being the most prevalent one. Among the 49 collected wigs, approximately 75% (37/49) were determined to be acrylics or modacrylics based upon microscopic observations alone. Fourier Transform Infrared (FTIR) spectroscopy was used to confirm the general polymeric class and further determine the subclass when possible. These data were useful in terms of studying the variation of the fiber types and subclasses within the sample population.

Wigs, Fibers, Trace Evidence

A128 Differentiation of Regioisomeric Alpha-Methyltryptamine (AMT; 3-(2-Aminopropyl)indole) and 5-IT (5-(2-Aminopropyl)indole)

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After attending this presentation, attendees will learn how a simple chemical derivatization and alternate GC/MS technique can be used to distinguish alpha-methyltryptamine (AMT; 3-(2-aminopropyl) indole) from its emerging regioisomer 5-IT (5-(2-aminopropyl) indole).

This presentation will impact the forensic science community by providing an optimized and simple chromatographic separation method for differentiation of the federally controlled substance AMT and the newly emerging substance 5-IT using N-methyl-N-(trimethylsilyl) trifluoroacetamide (MSTFA) with 1% Trimethylchlorosilane (TMCS) as a derivatizing reagent for Gas Chromatography/Mass Spectrometry (GC/MS) identification.

"Designer drugs" are created by altering the structural arrangement of currently controlled substances in an attempt to circumvent legal regulations, such as AMT and 5-IT. A dramatic increase in designer drugs has been observed in recent years, creating a significant analytical challenge. Regioisomers are particularly challenging to differentiate due to their structural similarities. The routine laboratory technique of GC/MS is often inadequate for the analysis of these types of compounds because they produce very similar or even identical Electron Impact (EI) mass spectra and may have similar or identical retention times.

Analysis of AMT and 5-IT using routine GC/MS conditions results in very similar retention times and EI fragmentation patterns for the two compounds. Therefore, these compounds cannot be unambiguously identified by this method alone. This presentation will describe the use of chemical derivatization in conjunction with routine GC/MS methods to distinguish of regioisomers. The use of alternative techniques such as Ultraviolet-Visible (UV-VIS) spectroscopy and various color tests will also be discussed.

A simple derivatization of AMT and 5-IT using MSTFA with 1% TMCS prior to GC-MS analysis results in two peaks for each compound, a mono-substituted product and a di-substituted product. While the MS fragmentation spectra remain equivalent for the derivatized AMT and 5-IT compounds, differing retention times allow for separation of the regioisomers. The mono-substituted products differ only slightly in retention time; however, the di-substituted products are well separated. Baseline resolution is not achieved under routine GC-MS analysis for the mono-substituted compounds, but it is achieved for the di-substituted compounds. The retention times and the MS fragmentation patterns are reproducible for the AMT and 5-IT compounds. The MS fragmentation spectra confirm the sample as one of the regioisomeric compounds, while the retention times identify the specific regioisomer.

Alternative methods of analysis not requiring derivatization were also investigated. Analysis of AMT and 5-IT using UV-VIS spectroscopy results in very similar spectra; however, the less intense, broader second peak did exhibit slight differences between AMT and 5-IT. Analysis resulted in a second peak at 273nm for AMT and 280nm for 5-IT. The broad nature of the peak and proximity of the peak apexes suggest that this form of analysis not be used for distinguishing the regioisomers. Analysis using both Ehrlich's and Marquis' color tests provided another method of differentiation between the regioisomers. The data obtained from analysis of pure analytical standards corroborated with the results reported by other studies. Unfortunately, when applied to unknown case samples, these methods did not provide the unambiguous distinction, presumably because of interference from the other components of the samples. These simple methods would be applicable only if the sample were pure.

The derivatization method using MSTFA had been successfully applied to several case samples indicating that regardless of impurities, samples containing AMT or 5-IT can be differentiated. This method can be easily transferred to other forensic labs equipped with GC/MS and can improve the identification and differentiation of samples containing these chemicals. The material included in this presentation is expected to be of interest to other

forensic chemists and laboratory personnel involved in the analysis and evaluation of controlled substance regioisomers.

Regioisomer, Designer Drugs, GC-MS

A129 Chemometrics Applied to 2D Raman Mapping of Trace Materials

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The goal of this presentation is to demonstrate applying multivariate statistical methods available in Renishaw WiRE 3.4 to interpret and validate Raman mapping data.

This presentation will impact the forensic science community by demonstrating the use of orthogonal statistical methodologies to corroborate analytical conclusions with the goal of encouraging forensic science laboratories to use statistical operations as part of routine analyses.

Chemometrics concerns the extraction of relevant information from chemical data using mathematical and statistical tools. In practice, it is the application of multivariate data analysis to chemical data.¹ Many chemometric methods rely on a wide variety of multivariate operations that are applied to chemical spectral data to separate noise from information, compare and classify spectra, and validate both methodology and conclusions. Although routinely applied in academic and research, forensic science laboratories do not routinely use chemometrics. In this report, spectral analysis is subjected to statistical methods that are part of the instrument software to demonstrate how to quickly verify and validate conclusions without using separate statistical software.

Maps are two-dimensional arrays composed of rows of locations called cells. The process is much the same as digital photography in which each cell is called a pixel. In both, each location contains information for that location and in the case of the mapped cell the information is both photographic and spectral. After data collection, the spectrum in each cell is identified by the statistical operations described below and is assigned a false color for identification. The map is then re-drawn using the false colors showing the location of the chemical components.

Mapping is particularly important in trace material analysis where the distribution and size of individual components are important identifying characteristics that can only be assessed by mapping the sample. Polymer identification, counterfeit pharmaceutical products, and ink location in altered documents are conventional forensic applications. Non-mapping analysis determines only a single or a small number of locations that, due to micro-heterogeneity, can lead to false identification. Mapping analysis corrects that deficiency.

Raman spectra were determined with a Renishaw In Via Raman microscope equipped with a motorized stage, and the data were analyzed using Direct Classical Least Squares (DCLS) component analysis, Principal Component Analysis (PCA), and Multivariate Curve Resolution (MCR) techniques available in Renishaw's WiRE[®] 3.4 instrument software.² The mapped data were evaluated by the three statistical methods with the goals of correct spectral assignment, and validation of one method by agreement with another.

The key to mapping analysis is the ability to accurately

identify the spectral information in each cell. DCLS relies on reference spectra for comparison and applies least squares analysis to compare the unknown spectrum to the reference spectra. MCR-ALS does not rely on reference spectra for comparison, but is able to extract component spectra from the data of a mixture. Usually, at least a single component of the mixture must produce a strong enough response to be unambiguously identified for this method to work. PCA does not produce spectral data, but reorganizes the data to groups (principal components) that account for the greatest variation. As such, PCA is often used to eliminate noise and sort spectra into groups having the same properties.

The presentation will include tables and colored maps that compare the statistical results.

References:

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Chemometrics, Spectral Mapping, Validation

A130 Characterization and Discrimination of Nail and Gel Polishes With Microscopy and Vibrational Microspectroscopy

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After attending this presentation, attendees will have a better understanding of the characterization and discriminating power of microscopic and spectroscopic methods for the analysis of nail and gel polishes.

This presentation will impact the forensic science community by characterizing nail and gel polishes and evaluating the discriminating potential of microscopic and spectroscopic methods for their analysis.

Nail polish is a very popular and potentially important type of cosmetic evidence because of its popularity, prevalence, and vast variety of colors, brands, and types available to consumers. Although not commonly analyzed by criminalists, nail polish identification and discrimination has played a role in criminal cases, including the infamous wood chipper murder. Nail polish is a quick-drying lacquer, either clear or colored, used to paint the fingernails or toenails. It contains multiple materials and is relatively complex. There are five main components in nail polish. The first is a film-forming material, which creates a protective coat on the polish. The second component is an adhesive material that helps hold the polish on the nail. The third is a plasticizer that makes the nail polish flexible instead of brittle. The fourth is a volatile solvent mixture, which holds all of the components together until the polish is applied to the nail. The solvent evaporates after application of the polish. The fifth ingredient is the color component which can be either organic dyes, inorganic pigments, or a mixture of both.

The purpose of this study was to analyze and characterize nail and gel polishes using microscopy and vibrational microspectroscopy. In this research, the classification and discriminatory power of these analytical methods was evaluated.

This study focused on the characterization and discrimination of both traditional nail polishes and the more modern gel polishes. For both types of polishes, only pink and red color

shades were included in the study. All of the colors chosen were as close in color as possible for the same brand as well as between brands. Seven different brands were chosen for each type of nail polish for a total of fourteen brands. From each brand, seven colors of similar shades were selected as samples, resulting in a total of 98 analyzed polishes. The brands used were a mix of salon quality and ones intended for at-home use.

This study used Raman and Fourier Transform Infrared spectroscopy (FTIR) microspectroscopy in addition to stereomicroscopy and polarized light microscopy to analyze the samples and gain information on their physical, optical, and chemical properties. Multivariate statistical analysis methods, such as principal component analysis and canonical variate analysis, were used to assess the discrimination ability of the Raman and FTIR spectroscopic methods. All 98 samples were able to be distinguished with the use of microscopy combined with vibrational spectroscopy.

Lastly, real-world nail and gel polish samples were analyzed to simulate actual samples that could be received in a forensic laboratory and to analyze the layer structure of the polish.

Nail Polish, Vibrational Spectroscopy, Microscopy

A131 Microscope Imaging of Cartridge Cases: Firearm Identification From Firing Pin Impression

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After attending this presentation, attendees will understand the main components of image analysis systems currently used worldwide by law enforcement agencies, investigators, and forensic scientists.

This presentation will impact the forensic science community by showing that further improvement of the identification of firearm evidence can result from the development of robust and automated computer algorithms applied to microscope imaging studies, as they provide quantitative and 3D information to the firearm examiner.

Firearms identification evidence — fired bullets and cartridge cases — provide important clues to solve criminal cases involving the use of a firearm. Forensic identification of ballistics specimens often relies on the detection, recognition, and matching of characteristic markings on their surfaces made by the firearms. This presentation focuses on microscopic image processing and statistical analysis techniques used to compare images of firing pin marks produced on the head of cartridge cases fired with different guns.

The presentation provides the description of a training image database of firearm evidence. The automated computer algorithms implemented for segmentation, registration, and classification of microscope images are described. A discussion of the strengths and weaknesses of the proposed comparison algorithm is given. After attending this presentation, attendees will understand the main components of image analysis systems, currently used worldwide by law enforcement agencies, investigators, and forensic scientists.

Further improvement of the identification of firearm evidence can result from the development of robust and automated

computer algorithms applied to microscopic imaging studies, as they provide quantitative and 3D information to the firearms examiner.

Depth from focus images (depth maps) and multi-focused planar images (textures) of characteristic marks were acquired for all evidence collected so far using a comparison microscope and stored in the database for further processing. For the evaluation of the comparison algorithm, available images of firing pins were examined. Cartridge cases (9mm Luger[®]) which were fired with two different hand guns/pistols (Glock[®] 17 and Walther[®] P38), with multi-focus images containing the entire firing pin mark, were examined. A training set of 30 digital images was found in the database, under the same image acquisition protocol: body part being examined (primer); lighting conditions (ring); imaging modality (multi-focus); and, level of magnification (20x).

Automatic pair-wise comparison of all selected cases is achieved in three main steps. First, all depth maps images in the training set are processed to extract the primer Region Of Interest (ROI). This segmentation stage is required to find the diameter and the center of the primer circle. Primer ROIs of equal size are extracted from all original images and translated to a common origin to obtain concentric primer circles. Image registration of a given pair of ROIs is then performed by step-by-step rotation. For each angular displacement, a normalized correlation metric is used to quantitatively measure how well one ROI compares to another. Optimal registration is achieved for the displacement that results in the minimum (optimal) metric value.

For the sake of classification, a dissimilarity-based classification strategy was adopted. Pairs of firing pin images were compared by a dissimilarity measure (derived from the optimal metric value computed in the previous step) reflecting their mutual resemblance. Therefore, for the given training set, a square matrix of pair-wise dissimilarities is produced, forming the dissimilarity matrix of all selected cases. Then, two samples (one for each gun) are selected for prototypes. The distance measure between the training sample and each prototype defines a new feature that when combined produces a 2D dissimilarity space. A linear discriminant classifier is finally built in this dissimilarity space. The six-fold cross-validation strategy is adopted for validating the methodology, associating correctly all the samples with the gun used for firing that evidence.

Preliminary results of recent investigations of the proposed comparison algorithm are presented. While growing, the database will be further investigated and other comparison pipelines will be implemented and tested.

Firearms, Imaging, Investigation

A132 Development of a Supported Liquid Extraction Method for Benzodiazepine in Urine With Surface-Enhanced Raman Spectroscopy Detection

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The goal of this presentation is to show the development of a Supported Liquid Extraction (SLE) method for the extraction of trace quantities of benzodiazepines in urine with Surface Enhanced Raman Spectroscopy (SERS) detection. The optimization of this technique as well as the limits of this method will be discussed.

This presentation will impact the forensic science community by detailing how this method has shown the

applicability of SLE for the efficient extraction of trace quantities of benzodiazepines from toxicological samples with SERS detection and the use of the technique over a wide range of compounds. SERS is more specific than currently used immunoassays because it provides spectral information for the compounds present.

Benzodiazepines are among the most frequently prescribed compounds for anti-anxiety and anti-depression and are commonly present in many toxicological screens. These drugs are also prominent in the commission of drug-facilitated sexual assaults due to their effects on the central nervous system. Due to their potency, a low dose of these compounds is often administered to victims; therefore, the target detection limit for these compounds in biological samples is 50ng/mL, which is well below therapeutic concentrations. Currently, these compounds are predominantly analyzed using immunoassay techniques; however, more specific screening methods are needed.

SERS has previously been shown to be able to detect trace quantities of benzodiazepines in aqueous solutions. This technique has the advantages of overcoming the low sensitivity and quenching the unwanted fluorescence effects seen with conventional Raman spectroscopy. SERS spectra are obtained by applying a compound of interest onto a SERS-active metal substrate such as colloidal metal particles or metal films. In this case, the colloidal particles are spherical gold nanoparticles in aqueous solution. SERS signals can be further increased with the addition of aggregate solutions. These agents are salt solutions which cause the nanoparticles to amass and form hot-spots which increase the signal intensity.

Spiked urine samples were prepared by adding diluted benzodiazepine and metabolite samples (prepared in 10% methanol) to drug-free urine at a range of benzodiazepine concentrations (1ng/mL-500ng/mL). An SLE method specific for benzodiazepines was used. Compared to the standard liquid-liquid extraction, SLE provides cleaner extracts and is less time consuming. A number of different method parameters were examined, including buffer concentration and pH as well as elution solvent. This extraction method has been shown to provide efficient extraction for the benzodiazepines in this study. Extraction efficiency was also determined. Aqueous colloidal dispersions of gold spherical nanoparticles were prepared using a modified Lee Meisel 1% sodium citrate reduction method. Particle size and shape were confirmed with an average size of approximately 30nm. Previous work has shown that for benzodiazepines, an aggregate solution made of MgCl₂ prepared at a concentration of 1.67M provided the highest signal intensity at the lowest drug concentration and was used in this study. Aggregate solutions were added to colloidal dispersions followed by the addition of extracted benzodiazepine samples and SERS spectra were obtained.

Overall, this method allows for the extraction and detection of a wide variety of benzodiazepines and their metabolites. The presence of individualizing spectral peaks provides a high degree of specificity for sample determination. The technique is sensitive with a limit of detection of 2.5ng/mL and linear over several orders of magnitude for the drugs chosen. This method has shown the applicability of SLE for the efficient extraction of trace quantities of benzodiazepines from toxicological samples with SERS detection and the use of the technique over a wide range of compounds. SERS is more specific than currently used immunoassays as it provides spectral information for the compounds present. Also, this technique has higher sensitivity and permits detection of drugs such as lorazepam, which have poor cross reactivity when using standard immunoassays.

Benzodiazepine, SERS, Drug Analysis

A133 Optimization of Headspace Solid Phase Microextraction Coupled With Gas Chromatography-Mass Spectrometry for Marijuana Profiling

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After attending this presentation, attendees will gain an understanding of Headspace Solid-Phase Microextraction (HS-SPME) methodology in order to profile trace samples of *Cannabis sativa L.*

This presentation will impact the forensic science community by providing preliminary results from a developed HS-SPME sampling method coupled with a Gas Chromatography/Mass Spectrometry (GC/MS) acquisition program for direct headspace sampling of cannabinoids and terpenes from solid samples of marijuana. This presentation will enhance the applicability of HS-SPME/GC/MS to controlled substance and trace evidence analysis.

Marijuana contains over 60 cannabinoids, of which the primary psychoactive component is Δ^9 -Tetrahydrocannabinol (THC). Current analytical methods of detecting cannabinoids and other constituents of marijuana include solvent extractions coupled with gas chromatography and liquid chromatography. Limitations of solvent extractions may include the use of harsh chemical solvents, expense, sensitivity, and time-consuming extraction steps. A proposed solution that corrects for these limitations is a modified HS/SPME/GC/MS methodology to detect and profile the cannabinoid and terpenic constituents found in marijuana plant material.

In this research, three cannabinoid and seven terpene reference standards (cannabidiol, THC, cannabinal, α -pinene, eucalyptol, 3-methyl-3-cyclohexen-1-one, (R)-(+)-limonene, myrcene, β -cedrene, α -caryophyllene), commonly seen in marijuana, were selected for the optimization of HS-SPME extraction condition. Mixtures of the standards were first injected in liquid samples onto the GC/MS in order to optimize the acquisition method. Following GC-MS acquisition optimization, mixtures of the standards were then prepared for the optimization of HS/SPME extraction. Before HS/SPME, the mixture of ten reference standards was prepared in GC vials. The solvent was dried under a gentle nitrogen stream before being capped and closed. The samples were then extracted using a Polydimethylsiloxane (PDMS) 24-gauge, 100 μ m absorbent fiber, testing for the optimal extraction temperature and time range for maximum recovery of standards with minimal interference. Optimum extraction temperature was found to be 95°C while the optimum time of extraction was ten minutes to prevent highly volatile compounds from being degraded. When HS/SPME samples under high temperature (95°C) with longer extraction time was tested (e.g., 30 minutes extraction), four of the seven terpenes (α -pinene, myrcene, limonene, and eucalyptol) were unable to be extracted. Under the optimal HS/SPME/GC/MS condition, as little as 11.1ng solid cannabinoids in a GC vial were able to be extracted and detected. Regeneration of the PDMS fiber was performed by exposing the fiber in a GC inlet under 280°C for 30 minutes to eliminate carryovers.

This study indicates that the proposed HS/SPME/GC/MS method is highly sensitive, able to detect cannabinoids in nanograms. Commonly seen chemical constituents in marijuana samples in their solid form could be extracted and detected using the optimal condition of HS/SPME/GC/MS method. Future research will include sampling of actual marijuana plant material in order to further optimize the extraction and acquisition parameters of the methodology for use in drug chemistry and forensic science laboratories.

Marijuana, HS-SPME, Drug Analysis

A134 Analysis of Portable Infrared Technology for the Identification of Solid Drug Samples

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After attending this presentation, attendees will understand the potential of handheld, portable infrared spectrometers in identifying and quantifying illicit drugs within a mixture.

This presentation will impact the forensic science community by evaluating the use of portable infrared instrumentation for faster, less expensive, and more efficient ways of identifying illicit drugs that could result in the reduction of laboratory backlogs.

The analysis and identification of illicit drugs constitutes the majority of casework performed in most forensic laboratories. This research evaluates the use of portable infrared spectrometers for the identification of suspected illicit drugs at crime scenes as a replacement for, or in addition to, the use of presumptive color tests. This has the potential to reduce the number of cases submitted to the controlled substance department of forensic laboratories. The recommendations by the Scientific Working Group for the Analysis of Seized Drugs (SWGDRUG) detail the combination of techniques that can be used for drug identification. When a Category A confirmatory method (such as infrared spectroscopy) is combined with a Category B or C method (such as a color test), then a drug can be positively identified. With the development of portable infrared spectrometers with sophisticated library search algorithms, this SWGDRUG protocol can be completed at the crime scene.

Using a portable infrared spectrometer as a tool for on-scene illicit drug identification is advantageous because it is reliable, easy to use, non-destructive, accomplishes analyses quickly, and creates a reviewable record of the results. New portable infrared instruments are small, lightweight, and can be safely and easily operated in extreme conditions such as could be encountered in clandestine drug laboratories or similar scenes. These portable infrared spectrometers utilize diamond Attenuated Total Reflection (d-ATR) sampling, thus no sample preparation is required beyond ensuring contact with the internal reflection element. Further, portable infrared spectrometers are equipped with sophisticated search algorithms that are specifically designed to determine if a sample is a mixture, and, if so, can identify and potentially quantify up to three mixture components. Additionally, these instruments are equipped with technology that enables the infrared data to be wirelessly transmitted to a computer command center for a permanent record of the data and more in-depth spectral analysis. In this research, several pure illicit drug samples, specifically cocaine, methamphetamine, MDMA, and heroin, along with two-

and three-component mixtures of these drugs, were analyzed with a portable infrared spectrometer to evaluate its use for the identification of illicit drugs and also for the quantification of illicit drug mixtures. Several samples of drugs commonly known as bath salts were also analyzed.

Infrared Spectroscopy, Controlled Substances, Drug Mixtures

A135 Scientist-Prosecutor Link to Reduce Forensic Biology Casework Backlog

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After attending this presentation, attendees will understand the method which is followed by the Forensic Biology (FB) section of Biological Inspection and Examination Branch in order to reduce the number of cases to be examined by discussion with the prosecutor in charge of the case.

This presentation will impact the forensic science community by reducing the forensic biology backlogs of cases through scientist-prosecution communication, cooperation, and case management.

The FB section covers the Emirate of Abu Dhabi with a few cases coming from other United Arab Emirates (UAE). The population of the Emirate of Abu Dhabi was 2.12 million in mid-2011, with an average annual growth of 7.7%. This indicates an increasing rate of population, which requires more effort on the part of the FB service. The FB section in the Abu Dhabi Police Forensic Evidence Department plays an important role in the examination, analysis, and recovery of biological evidence for several customers in the police stations and prosecution department, which numbers 41 agencies. The duties of the Public Prosecution Office (PPO) include filing the criminal cases, penitentiary, protective custody, and holding facilities. Forensic exhibits are sent to the FB section by the Crime Scene administration.

In many cases, the prosecution does not understand the background about these cases, as they could take longer to reach them due to administrative regulation. On the other hand, the case which was received could have been closed by the prosecution without informing the FB section. This can result in wasted effort expended in examining these cases.

Upon receipt, the exhibits are assessed and the case evaluated internally in the FB section based on priority and arrival time, and the cases which require a prosecution order to examine or to reject it will be identified according to certain characteristics. This will include the type of the case, the number of exhibits, and the objective of the examination. A scientist will meet the prosecutor who is in charge of the case in order to discuss the examination request. A case can be returned to the submitting authority if the prosecution rejects the examination request after the scientist-prosecutor meeting.

Several cases were removed from the FB section and returned after the prosecution decision that there was no need for examination. From 2010 to May 2013, there were 424 cases returned and not analyzed due to the prosecution order. The trend in the increased number of rejected cases: in 2010 there were 104 cases; in 2011 there were 115 cases; in 2012 there were 136 cases; and, through May 2013, 69 cases were rejected by the prosecution. There is a need for more research in the scientist-prosecutor link to increase the forensic examination performance and to reduce unnecessary casework.

Forensic Biology, Prosecution, Backlog

A136 Raman Spectroscopy as a Tool to Measure Laboratory Production Processes of *Bacillus Cereus* Spores

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After attending this presentation, attendees will understand the significance of the growth medium composition on the physicochemical properties (phenotypes) of bacteria, the forensic issues associated with microbial threat agent characterization, and the use of Raman spectroscopy with multivariate data analysis in forensic identification.

This presentation will impact the forensic community by not only introducing a rapid spectroscopic technique that can assist in the characterization of microbial evidence, but also by demonstrating how experimental repeatability and reproducibility affect the conclusion reached from the analysis. Because an organism's phenotype is influenced by laboratory culturing conditions, knowledge of the growth media signatures may help provide investigative leads and/or exclude a suspect facility during a forensic investigation.

Analytical procedures to determine signatures for production processes have been established using the analysis of stable isotope ratios, carbohydrates, fatty acids, and agar residues. Raman spectroscopy provides an attractive combination of rapid analysis with minimal sample preparation, non-destructive analysis, small required sample sizes, and low detection limits. Raman spectra are composed of energy-scattering bands due to vibrational movements of chemical bonds in nucleic acids, lipids, proteins, carbohydrates, and small molecules. As a result, this technique reflects the overall composition of the cell and, in turn, provides more information than the aforementioned analytical procedures.

Raman has been proven successful in the identification of bacterial species and strains. Changes in Raman spectra have been observed in the same species grown in different culture conditions; however, the repeatability of spore preparation using the same species of bacteria grown under the same culture conditions has not been fully investigated. Successful discrimination of bacterial spore production recipes requires signature bands specific to a species' metabolism of the culture medium; however, it also requires determination of the minimum within-source variability (variation in repeated preparations of bacteria under the same culture conditions) and its comparison to the between-source variability (variation between different spore production recipes).

The purpose of this research is to use Raman spectroscopy to identify reliable signatures due to the culturing conditions of bacterial spores. The research focused on growth of *B. cereus* T-strain (*BcT*) in five culture media (each with at least three independent preparations): Modified G (MG); Medium Brain Heart Infusion G (MBG); Schaeffer's sporulation broth and agar; and Columbia Agar with Blood (CAB) to determine the repeatability of Raman spectra of *BcT* in each of these five media.

To estimate the relative within- and between-source variability, Raman spectra were collected from multiple preparations of *BcT* spores grown in the various media listed above. For each medium, the mean spectra of the spores and the distribution of all individual spectra belonging to the media were used to represent the variation between preparations. The use of multivariate statistics accentuates subtle changes in the Raman spectra which, in turn, provide an indication of the reliability and reproducibility

of the bacterial spectra. Principle Components Analysis (PCA) followed by Linear Discriminate Analysis (LDA) with complete leave-one-out cross-validation was used to classify bacteria based on the Raman shifts that account for the most variability between each preparation.

Raman Spectroscopy, Multivariate Statistics, Microbial Forensics

A137 Evaluation of the Volatiles Organic Compounds Released From an Illicit Substance for the Development of Improved Field Calibrants for Instrumental and Canine Detection

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After attending this presentation, attendees will have an enhanced understanding on the importance of identifying the active odor signature of illicit substances and its use to aid in the development of field calibrants for detection by instruments and detection canines.

This presentation will impact the forensic science community by providing a detailed overview of the most abundant Volatile Organic Compounds (VOCs) that constitute the dominant odor profile of a frequently smuggled illicit substance and its potential in the development of instrument calibrants and canine training aids that are safer, easier to use, and provide a more reliable odor source.

Despite tough anti-drug laws enforced within the United States, the trafficking and use of illicit substances continues to be a national problem. The Office of National Drug Control Policy estimates that Americans spend approximately \$65 billion per year on illegal drugs from which only approximately \$1 billion (1.5%) is seized per year, domestically, by all Federal agencies combined.¹ Therefore, the detection of these substances, especially during transportation and storage, is of great importance and an emphasis is needed on improving current detection capabilities. Currently, canines are the most commonly used form of detection for concealed contraband. To maintain the operational readiness and reliability of these canines, routine training must be performed. Most often, detection canines are trained using gram quantities of illicit drugs, but there are challenges associated with this approach including needing to follow strict Drug Enforcement Administration (DEA) regulations, potential toxicities, and the variation in the amount of VOCs in drugs from different sources. As a substitute for real narcotics, odor mimics have been used for canine training purposes as they provide more consistent amounts of VOCs, are non-hazardous, and not subject to DEA restrictions.

Ongoing research has identified dominant active odor signatures of the major drugs such as cocaine, methamphetamine, and MDMA to include methyl benzoate, piperonal, and benzaldehyde, respectively, and effective odor mimics have been created using the identified active odor chemicals.²⁻⁴ Current research is focused on identifying the active odor signature of other popular illicit drugs, such as heroin. This study presents the VOCs released from heroin using Headspace Solid-Phase Microextraction Coupled with Gas Chromatography/Mass Spectrometry (HS/SPME/GC/MS) to identify the active odor signature. Depending upon the manner in which heroin is manufactured and sold, the constituents present may differ; therefore, headspace analysis

was performed on different batches of seized heroin and the VOCs detected were assessed for similarities and dissimilarities. Overall, over five different compounds were discovered and ranked by chemical functionality. A pilot study using narcotic detection canines was performed with odor mimics that were created using each individual compound, as well as mixtures, and the canines' response to each type of mimic will be presented. In general, the results demonstrate that HS-SPME-GC/MS of illicit substances combined with field testing with law enforcement-certified detection canines can be combined to develop reliable odor mimics for field calibration of detection canines which will result in an overall improvement in seizure rates of these substances.

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Illicit Substances, Volatile Organic Compounds, Detector Canines

A138 Does the Prior Application of the Field Kit BTK3 on a Suspected Bullet Hole Bias the Analysis of Atomic Absorption Spectrophotometry (AAS)?

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After attending this presentation, attendees will be informed regarding the application of field tests which do not bias the outcome of more sensitive analytical methods such as Atomic Absorption Spectrophotometry (AAS).

This presentation will impact the forensic science community by providing relevant information regarding forensic ballistics.

Forensic ballistics is the study of bullet trajectory and mainly consists of determining Gunshot Residue (GSR) to identify bullet holes. Firing a weapon produces a pall of discharge residue and traces left by smoke, dust, and fire at the moment of explosion, which is the result of combustion of both the primer and power of the cartridge. This so-called GSR can be found around the bullet entrance hole in cases of close- and medium-range shooting distances. The analysis of these residues around the entrance hole or wound by identifying individual morphological

and elemental characteristics enables the identification of GSR particles.¹⁻³ In particular, high concentration levels of three heavy metals — lead (Pb), barium (Ba), and antimony (Sb) — around a bullet hole provide grounds for assuming that the hole in question is an entrance hole.⁴

Both qualitative and quantitative analytical methods are available for the detection of GSR. Basically, in the laboratory highly sensitive methods such as AAS (also known as Atomic Absorption Analysis (AAA)), Inductively Coupled Plasma-Atomic Emission Spectrometry (ICP/AES), Scanning Electron Microscope (SEM-EDX), and Atomic Force Microscopy (AFM) are employed to analyze GSR.⁵⁻⁹

However, it is sometimes necessary to identify bullet holes immediately at a crime scene. This can be enabled through visualization techniques, so-called color tests, to identify bullet holes quickly (flash tests). Among others, the user-friendly “Bullet Hole Testing Kit 3” (BTK3) was generated to serve as a portable laboratory for the rapid identification of potential entrance holes by determination of the presence of GSR.¹⁰ Generally, these tests enable even the estimation of firing distances and direction from which the bullet was fired. The main purpose of this examination was to determine whether the implementation of the field test BTK3 on a suspected bullet hole would influence the outcome of AAS testing. Three commonly encountered firearms (two pistols, a Glock® 17 — caliber 9x19mm Parabellum with full metal jacketed lead core bullet, a Tokarev® — caliber 7.62mm Tokarev® with full-metal jacketed lead-core bullet, and one revolver, a Colt® Python® — caliber .357 Magnum® with semi-jacked lead core bullet) were fired at skin (pigskin) to simulate biological material, unpainted wood to simulate wooden structures such as doors, and cotton cloth to simulate clothing. All experimental shots were fired at a standardized angle (hit angle: 90°) at two different distances (30cm and 150cm), indoors in an adequately equipped laboratory setting. Subsequently, AAS-analysis was performed with and without previous BTK3 application.

In conclusion, the results clearly indicate that there is no significant interaction on the grounds of BTK3 use (BTK3 versus no-BTK3 (kit_nokit) (Pb: $p = 0.1309$; Sb: $p = 0.9111$), material*kit_nokit (Pb: $p = 0.5960$; Sb: $p = 0.9930$), distance*kit_nokit (Pb: $p = 0.4014$; Sb: $p = 0.9184$), and firearm type*kit_nokit (Pb: $p = 0.9662$; Sb: $p = 0.9885$)); hence, applying this field kit does not falsify later AAS outcomes.

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Bullet Hole Testing Kit (BTK3), Forensic Ballistics, Gunshot Residue (GSR)

A139 Error Rates for the Identification of 9mm Firearms Using the IBIS

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After attending this presentation, attendees will have a firm understanding of statistical methods as they relate to the analysis of firearms impression evidence. The generation of Receiver Operating Characteristics (ROC) curves will aid in the analysis of best-known nonmatches for 9mm firearms. The ROC curves will allow for a visual representation of false positive and false negative rates.

The presentation will impact the forensic science community by addressing current concerns of firearms examiners relating to data interpretation. The results will address the National Academy of Sciences (NAS) Report regarding the scientific basis of firearms examinations. The main benefits will be the development of a system based upon instrumentation and data which are commonly available in forensic laboratories.

According to the National Institute of Justice (NIJ), the degree of correspondence which must be exceeded in order to reach sufficient agreement to effect an identification is the Best Known Non-Match (BKNM) as determined by each individual examiner and as produced by different tools.¹ The individual examiner gains this experience during an initial training period rather than at the beginning of casework examinations. Anecdotally, it is known that examiners do find better BKNMs during casework. In order to understand this process, this concept will be elaborated upon.²

The Integrated Ballistic Identification System (IBIS) developed by Forensic Technology International serves as the backbone of the National Integrated Ballistic Information Network (NIBIN) system.³ This system allows for the data-basing of images of cartridge cases and bullets. For each cartridge case, two areas are imaged: the firing pin impression and the breech face impression. The proposed investigation involved the acquisition of 16 9mm center-fire handguns. All of the cartridge cases were entered into the 2D IBIS system in order to generate the match data. The acquisition method followed was established in a previous study.⁴ The data was mined to evaluate the variance within/ between relationships such as model, makes, firing conditions, etc.

One issue encountered in firearms is the determination of the number of Consecutively Matching Striations (CMS) method as described by Biassoti in 1959.⁵ There has been much debate between the so-called *pattern matchers* and *line counters*. According to Nichols, one of the pitfalls of the CMS method is the large degree of false exclusions.⁴ The discriminating ability of a method can be described by its sensitivity and specificity. The sensitivity of a method is its ability to detect a condition when the condition is present (or calling a *match*, a *match*). Specificity is

the ability of a method to detect an absence when the condition is not present (or calling a *non-match* a *non-match*). One can also define methods according to their false positive rate (*fpr*) and their false negative rate (*fnr*). The concept of a BKNM can be defined in terms of the *fpr*; as the *fpr* tends to zero, the *fnr* tends to one. Thus, irrespective of a CMS or pattern matching approach, the concept of false exclusions is unavoidable. Conversely, if one reduces the number of false exclusions, the *fpr* rate will increase.

The ROC curves can be used to determine the crossovers between match and non-match. The ROC curve demonstrates the discriminating power of the method. In other words, it determines how well the method can differentiate between different states of the samples to which the method has been applied. This discriminating ability is directly related to the area under the ROC curve. The error associated with this method is determined by the parameter under evaluation. The *fpr* and the *fnr* are given as a function of the correlation scores which were obtained by the IBIS. The crossover from black to gray to white zones are indicated when the error rates are zero. The gray is where the match and non-match scores overlap. It is in this gray zone where the quality-quantity relationship is the most critical. The firearms examiner must apply skills, knowledge, and experience to evaluate striations to be able to ensure a valid conclusion. The ROC curve will also provide a measure of method discrimination irrespective of the method which the examiner provided. The data generated will be transformed to develop likelihood ratios for the interpretation of firearms evidence.

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Firearms, IBIS, ROC Curve

A140 Chemical Profiling of Forensically Relevant Bacterial Threat Agents With Direct Analysis in Real-Time Mass Spectrometry (DART®-MS) and Fatty Acid Methyl Ester (FAME) Analysis

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After attending this presentation, attendees will understand how Direct Analysis in Real Time-Mass Spectrometry (DART®-MS) and Gas Chromatography (GC) can be used to analyze the surface composition of bacterial spore samples and how this can assist a forensic investigation.

This presentation will impact the forensic science community by introducing a novel strategy for nondestructively analyzing microbiological evidence. DART®-MS and GC techniques

can be integrated into existing operational workflows and can add new information to traditional molecular signatures (i.e., genotypic profiling) of microbial threat agents.

The ability to combine taxonomic information from bacterial threat agents with molecular signatures for the laboratory of origin has considerable implications for the forensic science community. Many mass spectrometry methods exist for collecting either type of signature; however, differences in sample preparation and sensitivity have prevented these techniques from being integrated into a comprehensive forensic method. Rapid chemical characterization techniques such as DART®-MS and GC are promising strategies to address these problems but have only been explored as tools for taxonomic characterizations.

The goal of this study was to test whether DART®-MS and GC could be used to collect molecular signatures from the spore surface that indicate both organism present and the growth medium formulation used during laboratory culturing. Two different strains of *Bacillus cereus* spores (T-strain and 14579) were cultured with three different medium formulations, each containing a different source of complex carbon/nitrogen compounds (tryptone, peptone, and yeast extract). Whole spore suspensions from each culture were analyzed with DART®-MS. Mass spectra were collected in the range of 50-1000m/z. In addition, chemical constituents of the spore surface were also extracted with an acetone wash and derivatized for Fatty Acid Methyl Ester (FAME) profiling using GC coupled to a Flame Ionization Detector (FID). Chemical profiles from DART®-MS and GC/FID were then compared.

DART®-MS results showed that mass spectral profiles of spores consisted of complex assemblages of both fatty acids and hydrophobic amino acids. The range of lipid structures included branched-chain odd, branched-chain even, and straight-chained fatty acids. The relative ratios of fatty acid structures were indicative of both strain and the culturing medium recipe. *Bc14579* spores were enriched in 17:0 iso fatty acids whereas *BcT* spores were enriched in 13:0 iso and 15:0 iso fatty acids. When spores of either strain were grown in tryptone formulations, DART®-MS profiles showed a higher proportion of 15:0 iso fatty acids whereas peptone cultures showed an increase in 14:0 iso and 16:0 iso fatty acids. Results from FAME profiles showed similar trends. The relative abundance of different fatty acid structure class was consistent with the ratios obtained from FAME extractions and GC/FID profiles of the same spore preparation. Individual FAME markers were found to differentiate *BcT* spores from *Bc14579*. For example, *BcT* showed a higher proportion of 17:1 iso ω5c (8% compared to 4%), while *Bc14579* exhibited a higher proportion of 17:0 iso (~19% compared to ~8%). When comparing the same organism grown on different medium formulations, it was observed that spores grown on tryptone were enriched in branched odd fatty acids (15:0 iso, 17:0 iso) and depleted in straight-chain fatty acids (14:0, 16:0, 16:1ω7c) compared to spores grown on peptone. Fatty acids 18:1 ω7c and 18:1 ω9c were abundant in the supernatant indicating their extracellular presence as opposed to intracellular. Taken together, these results suggest that DART®-MS and GC/FID can be powerful tools for taxonomic characterizations of unidentified spores and for the determination of forensically relevant aspects of the culturing procedure such as the source of complex nutrients.

Bacterial Spores, DART®-MS, FAME Profiling

A141 A Novel Mathematical and Digital Image-Based Approach to Gunshot Residue Pattern Interpretation

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After attending this presentation, attendees will understand how digital analysis and numerical interpretation can be applied to gunshot residue patterns. They will know the process by which the patterns were analyzed and the implications this method may have for crime scene investigations.

This presentation will impact the forensic science community by providing a possible method for mathematical support to a field that is largely dependent on subjective interpretation. It will also present a possible option for interpretation of gunshot residue patterns when the firearm in question is not recovered.

Traditional methods of Gun Shot Residue (GSR) pattern interpretation primarily rely on methods such as trial and error, shot re-creation, and sometimes chemical composition analysis in order to estimate muzzle-to-target distances. Most often in these investigations, the firearm in question must be test fired at different distances in order to create patterns to compare to the evidence. These methods rely on judgment and estimation without statistical or numerical support. In other cases, the actual firearm used in the crime is not even recovered to use for test firing. The goal of this study is to test an alternative method that may not only provide statistical support to a firearm/GSR pattern match, but may also aid in identifying or at least narrowing down possibilities for the gun that was fired.

In this study, a new technique for analyzing GSR patterns was preliminarily tested for its possible application to firearm distance and caliber determinations. Under controlled conditions, four different-caliber firearms were used to create GSR patterns, fired at distances ranging from six inches to sixty inches, onto white targets. These patterns were then digitally photographed and uploaded to the software program ImageJ, a program developed by the National Institute of Health that is used in photo analysis for various scientific applications. The photograph of each GSR pattern was analyzed in ImageJ by calculating the average gray value of different areas of the pattern, starting with a one-centimeter-square area directly surrounding the bullet hole (deemed "center" of the pattern) and progressing outwards in concentric squares to the edge of the target containing the pattern (up to 20 centimeters squared). These gray values were compared to the gray scale that was created in each picture using white and black reference squares photographed next to each pattern. The reference squares used were consistent across all photographs, but the gray reference scale was recalculated for each individual photograph in order to take into account lighting differences, a problem that would be very likely to occur in actual crime scene work. The result of these comparisons produced a "relative grayness" output value that could be translated into a mathematical measure of how much GSR was present in each measured region versus the amount of white space left untouched.

Several results were obtained from these initial studies. One such result is that there was a certain distance for each caliber at which there was not enough GSR residue that reached the target for it to produce any data using this analysis method; however, at the closer distances, preliminary analysis of the targets showed a clear trend of variation between the relative grayness gradient of GSR patterns at different distances for the same firearm. Although there was not enough data at this early stage to produce definitive statistics, using the limited samples, there appeared to be a variance of anywhere from one to three standard deviations

in some of the sequential distances tested with the same firearm. Additionally, each pattern was treated with either basic hairspray, Rust-Oleum® fixative, or no fixative, and there did not appear to be any significant difference in the stability of the pattern between the three methods.

This work has shown that this method of analysis has potential for application in real-case scenarios involving GSR patterns at close ranges. Information from further studies on this method may allow for this digital image-based process to introduce statistics and analytical methods to GSR pattern interpretation, both of which are virtually absent in present techniques.

GSR Pattern, Digital Picture Analysis, Firearm

A142 Validation of Liquid Chromatography Methods for Trace Analysis of Dyes Extracted From Acrylic, Cotton, Nylon, and Polyester Fibers Using UV/Visible and Mass Spectrometric Detection

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After attending this presentation, attendees will be knowledgeable about analytical methods for microextraction of dyes from trace evidence fibers. Dyes can then be identified by standard liquid chromatography with UV/Visible detection or structural characterization can be accomplished by mass spectrometry. Participants will also gain an understanding of the validation protocols which have been followed to document accuracy, precisions, and limits of detection.

This presentation will impact the forensic science community by demonstrating validated protocols for dye extraction from trace evidence fibers of 1mm. The profiling of dye formulations on trace fibers at parts-per-million (ppm) levels allows match exclusion conclusions to be made with higher reliability, and "results consistent with" will have increased probative significance.

Forensic fiber examinations involve comparison of trace evidence fibers to determine possible associations between victims, suspects, and crime scenes. Fiber evidence is class evidence; discovery of a fiber at a crime scene and its identification as a particular fiber type (e.g., acrylic, cotton, nylon, polyester) may not provide much support for a forensic investigation. What is required is information that makes the trace evidence more specific and discriminating. While fast nondestructive methods such as microscopy and UV/Visible microspectrophotometry or Infrared (IR) spectroscopy are preferred for forensic fiber examinations, these techniques do not always provide enough discriminative information to establish a common origin between fibers. The premise of ongoing research is that if dye formulations on trace fibers can be reliably profiled at trace levels, match exclusions can be made with higher reliability, and "results consistent with" will have increased significance. Population studies report that fibers at crime scenes can be as small as 2mm in length. Because extraction of dyes from a fiber is destructive to the evidence, it is desirable to analyze fibers in the mm or sub-mm range.

Fibers that are similar in visual appearance can often be discriminated by retention time matching and UV/Visible profile comparison without the need for Mass Spectrometry (MS). The use of tandem mass spectrometry offers provides sensitivity and unequivocal structural identification of the dye components.

Separation and detection of individual dye components provides a qualitative and semi-quantitative fiber dye “fingerprint.” Determining the number and relative amounts of dyes present, and characterizing those dyes at the molecular level by MS, offers an entirely new level of discrimination. Such information may also open the possibility of tracing specific dye formulations to the textile manufacturer.

Because dyes adhere to different polymer fibers with different mechanisms, extraction methods must be individually designed to disrupt those mechanisms and provide efficient extraction. For example, for nylon, an extraction solvent mixture of water, pyridine, and ammonia disrupts the electrostatic attraction of acid dyes to nylon; however, a single ultra-performance liquid chromatography method suited for qualitative and semi-quantitative analysis of all three dye types has been developed. Having a single chromatographic method for those three dye classes avoids using multiple chromatographic conditions and increases sample throughput.

Method transfer has been accomplished by the forensic laboratory of the South Carolina Law Enforcement Division (SLED). To maintain American Society of Crime Laboratory Directors/Laboratory Accreditation Board (ASCLD/LAB) accreditation, the Scientific Working Group for Toxicology (SWGTOX) validation guidelines were followed. SWGTOX requires accuracy within 20%, limit of detection and limit of quantitation of 10ng/mL or less, and a percent coefficient of variation less than 20% (within and between run). The method presented here for the three fiber types and each respective dye for Ultra Performance Liquid Chromatography — Diode Array Detection (UPLC-DAD) has resulted in a limit of detection of <570ppb, a limit of quantitation of <1.89ppm, and a coefficient of variation of <3.43% for all tested dyes. Although this approach is destructive to the fiber evidence, the ability to analyze sub-millimeter fiber lengths of single fibers, coupled with detection limits in the hundred picogram range by both DAD and tandem Mass Spectrometry (MS/MS), make routine forensic characterization feasible.

Fiber Analysis, Dye Extraction, UPLC and Mass Spectrometry

A143 Investigation of 2D-LC/QTOF/MS for the Detection of Illicit Drugs

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After attending this presentation, attendees will gain an understanding of Two-Dimensional Liquid Chromatography (2D-LC) technique coupled with Quadrupole Time-Of-Flight Mass Spectrometry (QTOF/MS). The attendees will understand how the technique can potentially be applied to resolving complex mixtures of forensically relevant samples in a single analysis.

This presentation will impact the forensic science community by discussing how the technique can be used to enhance data quality, allowing for an increased chromatographic peak capacity and sensitivity for complex sample matrices. The findings and methodology can provide forensic laboratories with the ability to identify alternative means for the detection and identification of seized drugs.

The majority of cases encountered by forensic chemists are analyzed utilizing traditional methodologies such as Gas Chromatography/Mass Spectrometry (GC/MS), Gas Chromatograph-Flame Ionization Detector (GC/FID), Fourier Transform Infrared-Attenuated Total Reflectance (FTIR-ATR), and Liquid Chromatography/Mass Spectrometry (LC/MS). The use of LC/QTOF/MS has gained popularity in recent years for its use in targeted screening as well as characterization of

unknown designer drugs. LC/QTOF/MS has a superior advantage over traditional methodologies as it can deliver accurate mass determination and provide highly specific mass-to-charge spectral data, with accuracies spanning in the milli-Dalton range for parent and fragment ions. As such, routine and targeted-based methods may be set up that are fully automated for screening and identification purposes. By integrating a second-dimension aspect to chromatography, the capabilities of the LC/QTOF/MS may further be enhanced, particularly for isobaric compounds or other designer drugs that are similar in structure, sharing some of the same chemical properties used to achieve chromatographic separation.

In order to investigate the suitability of 2D-LC/QTOF/MS for identification of seized drugs, experiments have been performed to assess operational characteristics in the analysis of 15 beta-ketone derivatives of amphetamine, commonly known as designer cathinones. The goal of this study was to assess the function of 2D-LC using an orthogonal combination of stationary phases, different mobile phase compositions, isobaric compounds, and limits of detection. The method employed an Agilent® 1200 binary pump with a Hydrophilic Interaction Liquid Chromatography (HILIC) column, coupled to an Agilent® 1290 Infinity Ultra High-Pressure Liquid Chromatography (UHPLC) (via a six-port valve) with a ZORBAX® Extend C18 column. TOF/MS full-scan spectra were acquired in positive Electrospray Ionization (ESI) mode over 50-1000m/z scan range using reference masses m/z 121.0509 and 922.0098. Additional TOF MS parameters were set as follows: fragmentor voltage at 150V; capillary voltage at 4,000V; skimmer voltage at 65V; nebulizer pressure at 50psi; gas temperature at 350°C; gas flow rate at 13L/min. Collision Induced Dissociation (CID) of precursor ions were obtained in targeted MS/MS mode using a collision cell with nitrogen as a collision gas.

Illicit Drugs, QTOF, 2D-LC

A144 Differentiation of Synthetic Cathinone and Synthetic Cannabinoid Regioisomers by GC-QQQ-MS/MS

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After attending this presentation, attendees will learn the use of Gas Chromatography Tandem Mass Spectrometry (GC/MS/MS) for the mass spectrometric differentiation of synthetic cathinone and synthetic cannabinoid regioisomers with both electron and chemical ionization modes.

This presentation will impact the forensic science community by providing the product ion scan mass spectra for the unequivocal identification of synthetic cathinone and synthetic cannabinoid regioisomers.

The abuse of designer drugs, especially for synthetic cathinones and synthetic cannabinoids, has proliferated worldwide with the ease of acquisition through the internet or head shops. Despite the efforts to regulate these psychoactive substances, new designer drugs, including isomers and derivatives, continue to emerge, replacing the controlled substances.¹ Gas Chromatography/Mass Spectrometry (GC/MS) is the gold standard analytical technique in many forensic laboratories as this technique enables the unambiguous identification of unknown compounds with the use of an Electron Ionization (EI) mass spectral library; however, the identification of designer drugs is often challenged due to their similar structures (derivatives) and the presence of positional isomers (regioisomers) resulting in very similar mass

spectral profiles in EI mass spectra and insufficient information for identification.² There have been many efforts to characterize regioisomers of amphetamine derivatives by mass spectrometric differentiation as well as chromatographic separation by GC/MS and Liquid Chromatography Mass Spectrometry (LC/MS).³ In this study, the mass spectrometric differentiation of several synthetic cathinone and synthetic cannabinoid regioisomers is performed by Gas Chromatography Triple Quadrupole Tandem Mass Spectrometry (GC-QQQ-MS/MS) with EI and Chemical Ionization (CI) modes. The discrimination of regioisomers is achieved by obtaining the product ion scan mass spectra at different collision energies, 10, 20, and 30eV. As a result of collision-induced dissociation of precursor ions, with nitrogen as a collision gas, the peaks for product ions are presented with different relative abundances in their mass spectra depending on the regioisomers. The advantage of using GC-QQQ-MS/MS as an alternative method for regioisomer differentiation over Nuclear Magnetic Resonance (NMR) spectroscopy is the ability to analyze trace amounts of sample. In addition, the obtained product ion scan mass spectra can be used as a supplemental library along with an EI full scan mass spectral database that is currently used in the library search for an unknown compound.

This approach will be presented for the analysis of regioisomers of 20 different synthetic cathinones and synthetic cannabinoids. For example, the regioisomers of AM694 were successfully discriminated with the characteristic product ion scan mass spectra at different collision energies.

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Synthetic Cathinones, Synthetic Cannabinoids, GC-QQQ-MS/MS

A145 Differentiation and Thermal Degradation of Synthetic Cathinone Regioisomers During Gas Chromatography/Mass Spectrometry Analysis

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After attending this presentation, attendees will learn about the thermal degradation of cathinones that occurs during Gas Chromatography/Mass Spectrometry (GC/MS) analysis and various methods to differentiate between regioisomeric compounds.

This presentation will impact the forensic science community by providing Electron Ionization (EI) mass spectral

data for the regioisomeric compounds and the presumed thermal degradation products of several synthetic cathinones, as well as some preventative and reductive measures related to this decomposition.

Synthetic cathinones, often referred to as "bath salts," are central nervous system stimulants and have been implicated in numerous fatalities.¹ Modifications to the aromatic ring, the alkyl side chain, and the amino group have all been observed for the synthetic cathinones. These compounds may be synthesized with ring substitutions at the *ortho*, *meta*, or *para* position to create new regioisomers for the controlled substance. This presents an analytical challenge in differentiating between some of the regioisomers, because the EI spectra and the retention times are often very similar using routine GC/MS analyses. Several methods with detailed mass spectral data will be presented to demonstrate methods for differentiation of 14 groups of regioisomeric synthetic cathinones, 35 total compounds. Adjustment of the method and chemical derivatization are the techniques used to achieve unambiguous identification of the regioisomeric compounds.

Thermal decomposition has been reported for some of these cathinones which was confirmed by data from this study.^{2,6} Stability of the regioisomers and their subsequent propensity for thermal decomposition were influenced by the positioning of the functional group on the aromatic ring for each group of regioisomers. The decomposition occurred in the injector port and/or the column, but the location varied between the groups of regioisomers. 2-Fluoromethcathinone exhibited unique decomposition products, supporting a previous study.^{2,6} Mass spectral data for the presumed degradation products will also be presented and the patterns observed regarding the decomposition location and functional group positioning will be discussed.

There is one major difference between data from this study and data reported previously by others. Others have found the use of splitless injection mode dramatically reduced the occurrence of thermal decomposition. In the present study, only split mode was used; however, increasing the split ratio actually increased the level of decomposition. This contrasted with the results observed in another study, but the change between ratios was greater in the present study and probably accounts for the difference.^{5,6} Other means for the prevention of thermal decomposition of these compounds have been determined by others and confirmed by the data from this study. Cleanliness of the liner and the use of a deactivated liner will substantially decrease thermal decomposition. Adjusting the GC/MS method may also decrease the thermal decomposition, as data indicate that some of the decomposition is occurring on the column; however, decreasing the temperature would also decrease the resolution and may prohibit the differentiation between the regioisomers of that compound.

The material included in this presentation is expected to be of interest to other forensic chemists, forensic toxicologists, and laboratory personnel involved in the analysis and evaluation of synthetic cathinone regioisomers.

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Thermal Decomposition, Cathinones, Regioisomers

A146 Microbial Degradation of Oxygenated Ignitable Liquids

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After attending this presentation, attendees will understand the concept of microbial degradation of the oxygenated ignitable liquids.

This presentation will impact the forensic science community by highlighting the major trends in bacterial degradation and recovery of oxygenated compounds in soil.

An organic-rich substrate such as soil is an excellent source of carbon for bacteria. Oxygenated compounds are also a rich carbon source, which can be utilized by bacteria. Previous work on the biodegradation of ignitable liquids has shown a significant loss of normal alkanes in the range of C₉ to C₁₆ as well as lower substituted aromatic compounds. This can be problematic for fire debris analysis as samples may sit for many weeks before they are analyzed due to case backlog. As a result, selective loss of key components due to bacterial metabolism can make identifying and classifying ignitable liquid residues by their chemical composition and boiling point range very difficult. Of particular interest in this study is to improve the recovery of oxygenated compounds by passive headspace concentration. Additionally, this study seeks to use this method to monitor bacterial degradation of these oxygenated ignitable liquids for inclusion in the Ignitable Liquids Reference Collection database developed by University of Central Florida and the Technical Working Group of Fire and Explosions to aid fire debris analysts in the identification of such liquids.

First, three different passive headspace methods were tested for the best recovery of the oxygenated liquids from Kimwipe® as well as soil. These methods included a 65°C bake for 16h, 85°C for 4h, and 85°C for 16h. The 85°C for the 4h method showed the highest relative abundance of the oxygenated liquids off of soil, so this is the passive headspace method that was chosen for the remainder of this study. To monitor degradation, 20µL of the oxygenated ignitable liquid was spiked onto 100g of potting soil and allowed to age for 0, 2, 7, 14, and 21 days. The samples were then analyzed using passive headspace adsorption followed by solvent desorption and then analyzed by Gas Chromatography/Mass Spectrometry (GC/MS). This study will show that within a few days the oxygenated compounds suffer significantly from microbial degradation while cyclic and branched alkanes present in these liquids are not degraded. Additionally, aldehydes present in the soil become apparent in the chromatograms of these liquids within as

little as seven days.

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Fire Debris, Oxygenates, Microbial Degradation

A147 Test Burns Using Clumps of Hair

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After attending this presentation, attendees will have learned about the effects of heat on masses of hair and the effects of surrounding materials on the microscopic appearance of the hair.

This presentation will impact the forensic science community by showing how the condition of hair at fire scenes is affected by surrounding materials and by stimulating work in this area. Interpretation of their microscopic appearance requires an understanding of the context in which hairs were burned. Coatings on hairs burned with different fabrics vary with the fabrics and the heating conditions.

In a study in which cotton T-shirts and blue jeans were set on fire and burned, clippings from the floor of a hair salon were wrapped in pieces of cotton knit paint rags and rayon fabric scraps and placed on an outdoor grill with burning blue jeans. At a later time, more hair clippings were rolled in T-shirt fabric, placed into a coffee can, set on fire, and allowed to smolder. In both cases, the appearance of the burned hairs was unexpected. Unlike individual burned hairs or smaller groups of burned hair, the wrapped hairs fused into a shiny carbonaceous network resembling a beaded bag in one case and a delicate network of fused fibers or threads in the other case. How much of the appearance could be attributed to the fabrics used to wrap the hairs and how much could be attributed to the behavior of the hairs themselves? What would happen if the hair were burned while still on a person or animal? What is the range of expected appearance and the variables that affect it?

Follow-up experiments were conducted with clumps of hair clippings and combings using the following steps: (1) placed into glass jars then heated on a gas stove; (2) placed into metal cans and heated on a gas stove; (3) placed in glass jars and metal cans together with a piece of paper lit with a match; (4) wrapped in cotton knit, cotton-polyester knit, rayon, cotton denim, thin nylon knit, silk, and polyester blouse fabric, all commonly encountered clothing fabrics; (5) placed into glass jars and metal cans then heated on an unlit grill; (6) wrapped in nylon knit and a thin cotton sock, placed in metal cans, and heated on a gas stove; (7) placed directly into a metal can and heated on a gas stove; (8) placed in a metal can and clumps of hair placed in a cotton sock, with the cotton sock and another sock wrapped around the can. The assembly was placed in a glass jar. The socks were ignited then allowed to smolder for several hours; (9) wrapped around a piece of pork then heated in a metal pan placed on a grill; and, (10) the resulting burned materials were examined using a stereomicroscope.

Portions of all the hairs exhibited swelling with a whitish appearance at low magnification; most are reddened and some are charred. The hairs that were heated in metal cans, whether wrapped or not, exhibited charring and melting. Hairs that were wrapped in silk were partly coated with melt and decomposition from the silk. Stove-heated hairs wrapped in polyester and nylon were initially protected while the fabric melted. When the gas flame was increased, raising the temperature, the hairs melted and charred, as did those on the grill.

Hairs wrapped in cotton charred and turned brittle when heated in cans on the stove, but when the cotton was ignited and allowed to smolder, exhibited the carbonaceous network and beaded appearance that was first remarked upon. The cotton itself, where distant from the hairs, exhibited black microspheres typical of cellulose fiber pyrolysis, but not as plentiful as those on the hairs. A sticky amber residue was observed on the sides of the burn jar. It is not yet known whether the tiny beads on the hair and the shiny coating are from the hairs themselves, from a pyrolysis product of cotton, or a combination. This question will be resolved with further testing.

Thermal Damage, Forensic Hair Exam, Fire Debris

A148 Quantitative Mapping of Post-Blast Nitroglycerin Residues on Pipe Bomb Fragments Using Total Vaporization Headspace (TV-HS) Solid Phase Microextraction-Gas Chromatography/Mass Spectrometry (SPME-GC/MS)

Dana Bors, 402 N Blackford Street, Indianapolis, IN 46202; and John V. Goodpaster, PhD, FIS Program, IUPUI, 402 N Blackford Street, LD 326, Indianapolis, IN 46202*

After attending this presentation, attendees will understand the importance of quantitation of explosive chemical residue on pipe bomb fragments, although it is not common investigative practice to do so. Quantitation will be accomplished using a new technique, Total Vaporization Headspace (TV-HS) Solid Phase Microextraction Gas Chromatography (SPME-GC/MS), which has several advantages over other methods. The validity of residue distribution mapping will also be discussed.

This presentation will impact the forensic science community by quantitation of explosive residue using a novel technique and diagramming its dispersal which can be valuable to both crime scene investigators and laboratory analysts examining pipe bomb fragments. By knowing the concentration of key residue components, analytical techniques can be tailored to optimize efficiency. This in turn will maximize throughput and evidence turnaround time. In addition, if the dispersal of residue indicates any trends, this information can heighten the understanding of the explosion process, such as the progression of deflagration.

Although residue from the explosive filler in a pipe bomb is routinely found on the post-blast container fragments, the amount of this residue is not quantified. The main reason for this is that the legal question at hand is what, not how much, explosive is present; however, there is value to tracking the distribution of explosive residue on device components. In particular, such "residue mapping" would provide general guidance as to what fragments may be more likely to contain high levels of residue. In addition, the distribution of residues would also shed light on the process by which the explosive filler deflagrates, resulting in the ultimate failure of the device container. In this study, TV-HS-SPME-GC/MS was used to identify low levels of nitroglycerin (~5ppb), which is an energetic component found in double-base smokeless powder. Traditional headspace SPME involves a three-phase system consisting of the sample, either in solid or liquid form, the headspace, and the fiber; however, by completely vaporizing the sample, the partitioning between the liquid and headspace is eliminated, simplifying the thermodynamics of the system and therefore increasing sensitivity. Another benefit of this technique is minimal sample preparation due to the high boiling, solid, and non-volatile components not transitioning into the headspace. Additionally, significantly higher sample volumes can be analyzed, even 100 times more when

compared to liquid injection. Ongoing work involves applying this analytical technique to numerous explosive devices constructed from Polyvinylchloride (PVC) and galvanized steel pipes and filled with Double-Base Smokeless Powder (DBSP).

Explosives, Nitroglycerin, SPME

A149 Forensic Analysis of Duct Tapes: Study of Scrim Via Scrim Count and Fiber Analysis

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After attending this presentation, attendees will learn of the forensic methodology adopted by the Forensic Chemistry and Physics Laboratory (FCPL) of Health Sciences Authority (HSA), Singapore, in the characterization of duct tapes and the characteristics of duct tapes commercially available in Singapore.

This presentation will impact the forensic science community by communicating that, other than legitimate uses, adhesive tapes are often used in numerous types of criminal activity such as homicide, kidnapping, and construction of improvised explosive devices. A majority of the cases submitted to FCPL involve using adhesive tapes to wrap drug packages for the purpose of illicit drug trafficking, which carries a serious penalty under Singapore's laws. For such cases, the value of adhesive tape examination is in associating strips of tape used to wrap different packages together or associating strips of tape from a wrapped package to a particular roll of recovered tape. Establishing such linkages could associate different suspects together or even seemingly independent cases to a drug syndicate.

One of the frequently encountered types of tape in casework is duct tape. Duct tapes generally consist of three layers: backing; fabric reinforcement (scrim); and adhesive. In this study, 50 rolls of duct tape that are commercially available in the Singapore market are characterized based on their physical characteristics (width, surface features on backing, and scrim count) and the type of fibers used in the scrim (twist, microscopic and fluorescence properties, melting range, cross section, and chemical composition). This study serves as a foundation to determine the frequency of occurrence of duct tape characteristics in the local market. The type and frequency of occurrence of manufacturing defects in the rolls of duct tapes studied will also be reported.

In the characterization of scrim, FCPL adopted the method derived from "Forensic analysis and discrimination of duct tapes," which described the scrim count per inch of duct tapes method (henceforth referred to as Method A).¹ Method A is useful in most cases except when the case involves small tape fragments or when the tapes are pasted on other types of evidence. Thus, FCPL explored two other methods (henceforth referred to as Methods B and C, respectively). Method B investigated the possible correlation between the surface characteristics on the backing and the warp yarns of the scrim. This method could potentially be useful in cases where the scrim is inaccessible for examination. Method C investigated the average length per warp and fill spacing to characterize fragments of tape.

*Presenting Author

The Discriminating Power (DP) of Methods A, B, and C as well as the respective combined DP with fiber analysis was determined to assess their usefulness, which could be particularly significant in situations where no physical fits are found. From this study, a general workflow to characterize duct tapes was implemented, based on their state and condition in which they are submitted.

Reference:

1. M J Bradley & A H Mehlretter. Forensic analysis and discrimination of duct tapes. JASTEE, vol. 3(1), pp.2-49 (2012).

Duct Tapes, Scrim Count, Fiber Analysis

A150 A Standard Method for Collection of Dichroic Spectra of Dyed Fiber Evidence

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The goal of this presentation is to help attendees learn, retain, or implement a method for the collection of dichroic spectra of dyed fiber evidence that addresses several factors that are necessary in order to obtain reproducible and reliable absorption spectral data from microscopic fiber evidence.

This presentation will impact the forensic science community by showing attendees that observing dichroic behavior and measuring dichroic spectra are essential discriminating factors in dyed fiber comparisons. Dyed fibers are a common class of trace evidence and microspectrometry is used to obtain absorption spectra for comparison of a questioned fiber to a fiber from a known source. This study identifies factors that must be considered when making dichroic spectral measurements. Failure to make dichroic spectral measurements can lead to false exclusions or excessive spectral variations which can lead to either false inclusions or cause the analyst to believe the spectra are unreliable. This refined method for dichroic spectral measurements improves the reliability of spectral data and provides significant additional information for comparing dyed fiber evidence.

Polarization is a commonly ignored variable in micro-spectral analysis of dyed fiber evidence. Since all textile fibers are oriented molecular structures, measuring the spectra of dyed fibers requires the analyst to control the state of polarization when recording their absorption spectra. In recent studies, it has been shown that a majority of dyed textile fibers are dichroic.¹ The absorption spectrum of a dichroic fiber is dependent upon the orientation of the light's electric field vector relative to the fiber's principal axes. Spectra must be recorded with linearly polarized light. The absorption spectrum of the fiber must be recorded with the electric field vector parallel to the longitudinal fiber axis and perpendicular to this fiber axis.

Grating and prism-based dispersive spectrometers produce a degree of polarization. This intrinsic polarization can be detected by recording the change in intensity when a linear polarizer is rotated through 360°. If the spectrometer has no intrinsic polarization, then the intensity will be constant as the linear polarizer is rotated. Using a spectrometer without knowing its intrinsic polarization can lead to serious problems. Placing a single linear polarizer either before or after the sample provides the necessary control to orient the radiation's electric field to the principle directions of the fiber. Since most micro-spectrometers operate in a single-beam mode it is necessary to record a background for each

orientation of the polarizer. The intrinsic polarization of several commercial micro-spectrometers and various dispersive elements will be presented to illustrate the magnitude of this problem. There is a mistaken belief that if a polarizer is not used, then there is no dichroic effect. The intrinsic polarization of the spectrometer is introducing an unseen variable when measuring the absorbance spectrum of a dyed fiber.

Reference:

1. K. De Wael, T. Vanden Driessche, Dichroism measurements in forensic fibre examination Part 1 - Dyed polyester fibres, Science and Justice 51 (2011) 57-67

Microspectroscopy, Dichroism, Intrinsic Polarization

A151 Rapid Determination of Adulterants in Aqueous Mixtures by DART®-MS

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After attending this presentation, attendees will understand how Direct Analysis in Real Time Mass Spectrometry (DART®-MS) can be used to screen for the presence of common adulterants in aqueous mixtures. Additionally, attendees will understand how the inclusion of a dopant can aid in obtaining better specificity in the identification of these compounds.

This presentation will impact the forensic science community by providing a method for the rapid screening of liquids to determine the presence or absence of common adulterants without the need for sample preparation.

DART®-MS is a rapidly growing technique in forensic science. In comparison to traditional mass spectrometry-based techniques, such as Gas Chromatography Mass Spectrometry (GC/MS), DART®-MS provides the benefits of analysis under ambient conditions without the need for sample preparation. Additionally, sample analysis can be completed in a number of seconds, unlike GC/MS which typically takes several to tens of minutes. However, the pitfall of DART®-MS is the potential lack of specificity in chemical identification due to a lack of chromatography or other separation technique. In complex mixtures, where a number of chemicals and their fragments are present, complex and ambiguous spectra are obtained, making it difficult to ascertain whether low weight peaks are individual chemicals or merely the fragments of larger molecules. This lack of specificity can, in part, be mitigated through the use of a dopant compound, which can provide characteristic adduct ions.

The application of DART®-MS discussed here deals with the detection of adulterants in aqueous mixtures, namely beverages. Detection of adulterants in beverages is necessary in a number of different cases including: homicide; attempted homicide; sexual assault; suicide; and domestic violence. Common adulterants in beverages include alcohols (such as methanol and isopropanol), acetone, ammonium, hypochlorite-based bleach, and radiator fluid (ethylene glycol). Detection of these compounds in beverages can be difficult due to their low molecular weight — which can make detection against background difficult — and high volatility.

In this study, a number of adulterants, including those listed above, were analyzed by DART®-MS, both in aqueous solutions and in complex mixtures. An optimized method for the detection of the compounds was determined, and it was found that this method provided limits of detection at 0.1% by volume or lower. The limits of detection exceeded that of headspace GC/MS, which is the traditional technique commonly used to analyze these samples. Furthermore, it has been found that these chemicals

can be detected in a number of different beverages, including sports drinks, sodas, and energy drinks. The use of dopants is also employed to allow for the determination of specific compounds which have similar fragmentation and dimerization products. The use of these dopants has been shown to differentiate the structural isomers propanol and isopropanol in both pure aqueous solutions and in beverages. Current work is being completed for the evaluation of a mixture of adulterants in solution as well as the detection of adulterants in ethanol-containing beverages.

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DART®-MS, Adulterants, Chemical Analysis

A152 Getting Past First Bayes With DNA Mixtures

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After attending this presentation, attendees will better understand the scientific foundation that underlies the DNA match statistic. Common misconceptions, and ways to avoid them, will be discussed.

This presentation will impact the forensic science community by enabling practitioners to more confidently present accurate DNA mixture results in the classroom and in court.

DNA mixtures are a prevalent form of biological evidence. A mixture contains DNA from two or more contributors. There are usually multiple genotype explanations for the observed Short Tandem Repeat (STR) data. Forensic scientists must understand genotype mixture inference in order to give accurate DNA mixture testimony in court.

Fortunately, Bayes theorem provides a robust framework for genotype inference and match. Over 250 years ago, the Reverend Thomas Bayes showed how to update the belief in hypotheses (probability) by examining how well those hypotheses explain observed data (likelihood). Bayes has individuals use all the data, and consider all hypotheses.

Bayesian genotype inference (for each contributor at every genetic locus) begins with a *prior* belief that the chance of observing an allele pair before seeing data is proportional to its population prevalence. Careful examination of STR data then uses a *likelihood* function to concentrate probability on those genotype values that best explain the laboratory data. This objectively inferred genotype associates a *posterior* probability with every allele pair, multiplying prior probability and likelihood.

A DNA match statistic assesses the strength of match between evidence and reference genotypes, relative to coincidence. This Bayesian Likelihood Ratio (LR) weighs two competing hypotheses — either the reference individual contributed DNA to the evidence, or he did not — based on the observed STR data.

Bayesian beginners often make mistakes. Beginners may fail to use all peak data, or not consider all genotype hypotheses. Bayesian beginners can confuse likelihood (chance of data given hypothesis) with probability (chance of hypothesis given data). A beginner may apply complex formulas when a simple ratio would suffice. Bayesian beginners may change assumptions in mid-step and suggest meaningless comparisons.

On April 12, 2013, the National Institute of Standards and Technology (NIST) Applied Genetics Group gave a full-day webinar

on DNA mixture interpretation. The NIST group presented genotype and LR results from Bayesian software. While attendees correctly computed the final LR match numbers, some beginner errors were made. DNA interpretation missteps that appear harmless in a pedagogical setting can prove damaging in a court of law, where accuracy is paramount and cross-examination unforgiving.

This presentation reviews the basic principles of Bayesian DNA mixture interpretation. The NIST webinar experience provides teaching points that can help beginners avoid common mistakes. The corrections provided to NIST after the webinar are used here to highlight key interpretation steps. With some Bayesian training, DNA analysts can accurately testify about mixture results and get past first Bayes.

Bayes Theorem, Genotype Inference, Likelihood Ratio

A153 Suspect-Centric Combined Probability of Inclusion: A Means of Attaching Objective Statistical Weights to Mixed DNA Profiles Where Dropout May Have Occurred

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After attending this presentation, attendees will have a better understanding of the difficulties associated with assigning a statistical weight to a mixed Short Tandem Repeat (STR) DNA profile where allelic dropout may have occurred. A solution to the problem that generates a suspect-centric combined probability of inclusion that can be empirically evaluated will be described.

This presentation will impact the forensic science community by providing a means by which statistical weights can be attached to mixed samples where allelic dropout may have occurred. A suspect-centric combined probability of inclusion can be evaluated by comparing the result of using the suspect's profile to results obtained from similar analyses with the profiles of a large population of individuals who could not have contributed to the mixture.

Attaching statistical weight to samples with contributions from more than one individual where allelic dropout may have occurred is one of the greatest difficulties facing forensic DNA profiling. Despite effort by numerous parties, there is still no generally accepted means of attaching a reliable statistical weight to such a result.

Generally speaking, there simply may not be enough information present within an STR DNA profile to allow a reasonable inference to be made regarding the likelihood of allelic dropout. At the very least, determination of allelic dropout rates from features of evidence samples themselves (such as relative peak positions and heights) is challenging and can lead to a range of estimates that dramatically impact the final statistical weight attached to a sample. The lower precision of quantitation results for low-level samples and difficulties determining the relative contributions and even the number of different contributors pose problems for alternative means of determining allelic dropout rates. Some testing laboratories opt, in error, to generate statistical weights for mixed profiles where allelic dropout may have occurred by simply disregarding information from loci where not all of a possible contributor's alleles are detected. Such a suspect-centric approach is biased and fails to take into consideration information that might support alternative theories of a case. As a result, work to develop a means to attach statistical weights to mixed samples where allelic dropout may have occurred has consciously endeavored to minimize the impact of allelic dropout and avoid using information

from a possible contributor's reference profile to resolve ambiguities associated with evidence samples.

Somewhat paradoxically, a suspect-centric approach that liberally invokes allelic dropout in a fashion that is heavily biased so as to include an individual as a possible contributor may offer an easily implemented and readily understood means of attaching a statistical weight to mixtures where allelic dropout may have occurred. Such an approach could be used to generate a "Suspect-Centric Combined Probability of Inclusion", or "SCCPI", using the standard CPI calculation for a mixed-evidence sample — but only for loci where all of a possible contributor's alleles are observed. The resulting statistic can then be directly compared to values similarly calculated for a large population (either real or simulated) that only contains individuals who could not have been a contributor to the mixed sample. If, for example, a given suspect is found to have a smaller SCCPI statistic than that found for any of 1,000,000 individuals from a relevant reference population, then that observation can be used to objectively support the proposition that the subject of the investigation is very likely to be a contributor to a mixture even when numerous instances of allelic dropout need to be invoked. In contrast, if a large fraction of 1,000,000 individuals from a reference population are found to have smaller SCCPI values than a possible contributor, then the possible contributor's similarity to a mixed DNA profile could be shown to be relatively unremarkable.

The nature of the distribution of SCCPI values for simulated individuals in large reference populations changes for different types of mixed DNA profiles. Evaluation of the distribution of reference SCCPI values for a mixed STR DNA profile even in the absence of reference samples from possible contributors may by itself provide a useful means of characterizing a sample's potential probative value in a way that could assist in prioritizing the further analysis of evidence samples.

Mixed, DNA, Dropout

A154 Using Simulation to Improve Understanding of Likelihood Ratio Results

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After attending this session, attendees will understand issues involved in presenting Likelihood Ratios (LRs) to non-scientists, such as judges and juries, and will see how simulation can be used to help explain the strength of an LR.

This presentation will impact the forensic science community as communication of results in court is an integral part of a DNA analyst's job. The topic should be of broad interest since many laboratories use LRs and wrestle with proper presentation of results in court.

An LR can be used to assign a statistical weight to a comparison between a DNA mixture obtained from an item of evidence and the DNA profile of a known individual, such as a suspect. The Office of Chief Medical Examiner (OCME) of New York City computes LRs using an in-house program, the Forensic Statistical Tool (FST), which models allelic drop-out and drop-in. This is the recommended approach for analysis of complex DNA mixtures by the DNA Commission of the International Society of Forensic Genetics.¹

To help interpret LRs, a qualitative scale of "limited," "moderate," "strong," or "very strong" support for one hypothesis over the other is used. Even with these qualifiers, it may be difficult for jurors to understand the strength of a result. In addition, the weight may vary depending on the model used, for example two-person versus three-person models, or with or without including a known contributor in the model. One piece of information that may be helpful in understanding the strength of a result would be the probability of obtaining such a result if the suspect is not a contributor to the mixture. In other words, what is the chance that a randomly chosen non-contributor would generate an LR at least as high as the one obtained for the suspect? Simulation and testing of non-contributor profiles can be used to estimate such a probability for a given result with a specific mixture. Others have explored this idea, using simulated profiles to show the distance between a suspect-generated LR and a set of non-contributor LRs.²

Estimating the probability of obtaining an LR at least as high as the suspect's requires that one or more simulated non-contributor LRs meet or exceed the LR computed for the suspect. With a low suspect LR, analysis of several thousand non-contributors may be sufficient. But, with a high suspect LR, several million non-contributors may be required in order to find any with LRs in the range of that of the suspect. While millions of non-contributor profiles can be simulated in seconds, the time required to perform LR analysis of those profiles may be a limiting factor. As a solution, many non-contributor profiles can be simulated and those with the fewest alleles missing from the casework mixture can be identified. These represent the non-contributors most likely to generate high LRs.

For example, with one two-person mock casework sample analyzed, one "suspect" generated an LR of 8.5, which would constitute limited support for the suspect, rather than an unknown, unrelated person, contributing to the mixture. Simulation and testing of 10,000 non-contributors yielded two profiles that generated LRs of 8.5 or more. Thus, the probability of obtaining an LR of at least 8.5 for this mixture is about one in 5,000. Using the same sample, another "suspect" generated an LR of 750, which would be classified as strong support for the scenario involving the suspect, over the scenario without the suspect. One million non-contributors were simulated and the top ten thousand were identified, five of which generated LRs greater than 750. Thus, the estimated probability of obtaining an LR of at least 750 for this mixture is estimated at one in 200,000.

The relationship between the number of missing alleles and the LR can be used to determine the appropriate cutoff value such that no high LRs are missed, but the number of profiles to be analyzed is within the range of feasibility. Regression analysis to determine the appropriate cutoff value for the number of missing alleles will be discussed. The cutoff may depend on the number of contributors to the sample and the probability of drop-out used by the LR program. Non-contributor results from analysis of a variety of mock casework mixtures will be presented and ideas for communication of results in court will be discussed.

References:

1. Gill P, Gusmao L, Haned H, Mayr WR, Morling N, Parson W, Prieto L, Prinz M, Schneider H, Schneider PM, Weir BS. DNA commission of the International Society of Forensic Genetics: Recommendations on the evaluation of STR typing results that may include drop-out and/or drop-in using probabilistic methods. *Forensic Science Int: Genet*, 2012;6:679-688
2. Gill P, Haned H. A new methodological framework to interpret complex DNA profiles using likelihood ratios. *Forensic Sci Int: Genet*, 2013;7:251-263.

Likelihood Ratio, DNA Mixtures, Simulation

A155 How Y Haplotypes Are Distributed and How They're Not

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After attending this presentation, attendees will have a mental picture of the patterns of groups of men who share Y haplotypes. This understanding is important for giving a realistic appraisal of the evidential strength of a Y haplotype forensic match.

This presentation will impact the forensic science community by simplifying and clarifying how to understand the evidential value of haplotype evidence — mtDNA as well as Y. It will not solve the problem of coping with population inhomogeneity, but will provide promising illumination for the way forward.

Forensic evaluation of DNA evidence rests in part on understanding the underlying population genetics — how the DNA is distributed in the population. This is especially true for lineage markers including Y haplotypes. It is natural, but wrong, to suppose that the familiar autosomal tools and principles adapt to the Y domain; they don't. Obviously, the autosomal product rule is lost, and there are important but less obvious examples as well. In particular, while sample frequency is a fair approximation to autosomal allele matching probability, for today's Y haplotypes of many STR loci, it's absurd. The common expression "unrelated man" is sometimes a good enough approximation for traditional work; however, since a patrilineal connection without intervening mutation is almost the only possibility for two men to share a Y haplotype, reasoning by analogy with autosomal DNA is to dramatically misperceive the evidence. The random mating/product rule approach to autosomal calculation using allele probabilities can be refined by taking into account the "inbreeding coefficient" θ , the chance two alleles have a common ancestor without mutation. The Y haplotypes situation is the opposite where one starts with θ and it is 97% of the story (for Yfiler®) and anything else is a minor refinement.

Alleles at neighboring autosomal STR loci replenish one another by step-wise mutation as is obvious from observing the compact and sometimes unimodal distribution of allele frequencies. It's natural to suppose that haplotypes behave similarly, but they don't. Mutation along a patrilineage away from a type, then mutation back to the original type is an insignificant phenomenon. To a close approximation, all the men in the population with any particular haplotype are men descended from a common ancestor (typically 500 years past) without mutation. The size of such a group is typically thousands or tens of thousands of men, all mutual cousins. Understanding how those cousins are distributed in the world is how to understand haplotype evidence.

Y-Haplotype, Lineage Marker, Population Genetics

A156 Characterization of DNA-Based Certified Reference Materials With New and Emerging Technologies

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After attending this presentation, attendees will understand the principles of digital Polymerase Chain Reaction (PCR), high-throughput sequencing, Sanger sequencing, and fragment-based

Short Tandem Repeat (STR) typing for the characterization of DNA-based reference materials used in the forensic DNA typing community.

This presentation will impact the forensic science community by detailing the techniques and protocols used for characterizing National Institute of Standards and Technology (NIST) Standard Reference Materials. The reference materials are commonly used for the calibration of STR typing methods and sequence determination, in addition to quantitative PCR in forensic laboratories.

Over the past 20 years, the Applied Genetics group at the U.S. NIST has been providing DNA-based Standard Reference Materials (SRMs) for the human identity testing community. Current examples include: SRM 2391c for STR typing; SRM 2372 for human DNA quantitation; and SRMs 2392 and 2392-I for mitochondrial sequencing DNA analysis.

SRM 2391c is required by the Federal Bureau of Investigation (FBI) DNA Quality Assurance Standards (Standard 9.5) to calibrate DNA typing procedures performed in forensic laboratories. SRM 2391c consists of genomic DNAs (≈ 100 ng in 50 μ L) that have been highly characterized for forensically relevant markers such as core autosomal and Y chromosomal STRs. The characterization of the forensic STR markers of interest is typically a combination of either Sanger sequencing or fragment size analysis. With the upcoming addition of new core autosomal STR loci and supplemental loci contained in commercial Y STR kits, updated information will be added to the characterization of SRM 2391c.

SRM 2372 is typically used for assigning a concentration to a working DNA standard for quantitative PCR measurements. Accurate quantitation of casework DNA extracts is important for determining optimal input amounts for the downstream PCR amplification of STR markers. The three components of SRM 2372 have been certified for Decadic Attenuance (Absorbance) in the single stranded state. This allows the assignment of a working informational value for each component in nanograms per microliter (ng/ μ L) (by assuming that one OD at 260nm equals 37 μ g/mL). Moving forward, using digital PCR methods to associate "copy number" characterization to nanograms per microliter concentration may prove even more useful in certain situations.

SRM 2392 and 2392-I contain extracted human DNA that has been characterized for the entire mitochondrial genome ($\approx 16,569$ base pairs) by Sanger sequencing methods. Multiple strands of coverage in both the forward and reverse direction across the entire mitochondrial genome ensures confidence in the base calls.

With the emerging application of High-Throughput Sequencing (HTS) and digital PCR technologies to genome characterization, NIST is starting to explore deeper characterization of forensic DNA-based SRMs. The considerations for additional characterization of reference materials include: source of genomic DNAs; amount of DNA required; genetic markers to be typed; analysis by multiple technology platforms; and the specific needs of the forensic community. This presentation will review the past SRMs and identify requirements for the analysis of future forensic reference materials. Examples of HTS and Sanger sequencing of SRMs 2391c and 2392 will be presented along with initial digital PCR results for copy number determination of SRM 2372. These emerging technologies often enable a more accurate assessment of the existing SRMs and provide added value to the forensic community.

PCR, STR, Mitochondrial

A157 Forensic Exploration of Temporal and

STR forensic identification is a powerful technique that depends upon collecting homogeneous genetic samples from a crime scene for Polymerase Chain Reaction (PCR) amplification; however, real-world samples often present mixtures of cells from different individuals at relatively low concentrations, and thus are difficult to type by conventional methods. Recently, the development of microfluidic technology has enabled digital PCR amplifications in ultralow-volume droplets with single template copy sensitivity. A microfluidic-droplet based approach was developed to perform digital STR typing of single cells with high sensitivity, fidelity, and throughput.

The overall strategy is to isolate individual cells in nanoliter droplets along with primer conjugated beads, release the nuclear DNA for STR amplification, and then analyze the single-cell amplicons linked to each bead. First, a microfluidic droplet generator was constructed with polydimethylsiloxane soft lithography that produced 1.5nL monodisperse agarose microdroplets in fluorinated oil at a high frequency of 444Hz. Second, single cells, along with microbeads functionalized with multiplex primers for STR targets, were dispersed in agarose and compartmentalized within the droplets at a statistically dilute level. The 34µm Sepharose® beads serve as amplicon-binding substrates during PCR to maintain the monoclonality of STR analysis. The unique thermo-responsive sol-gel switching property of agarose enabled the gelled droplets containing the individual cells (and beads) to be processed for various mechanical manipulations and storage while preserving single-genome integrity. Third, the porous structure of the gelled agarose was exploited to introduce a lysis and digestion buffer which released high-molecular-weight genomic DNA from the cell but kept the template DNA entrapped within the droplet. Fourth, the gelled droplets were equilibrated with PCR mixture and redispersed in silicon carrier oil by agitation to form a uniform emulsion of nanoliter PCR reactors. Massively parallel single-cell emulsion PCR was then performed in a standard PCR tube using a conventional thermocycler. This process transferred the STR loci information from the trapped cell onto the co-encapsulated microbead within the droplet. Following amplification, the beads were recovered by removing the oil and disrupting the droplets by melting the agarose. Finally, the STR products immobilized on the beads were detected by reamplifying in PCR plates under appropriate statistically dilute conditions followed by conventional Capillary Electrophoresis (CE) fragment analysis.

The utility of this method for forensic DNA typing was first explored using a 9-plex STR system employing eight core STR loci in the Combined DNA Index System (CODIS) database (D3S1358, D5S818, D7S820, D8S1179, D13S317, D21S11, vWA, and TH01) plus the sex marker Amelogenin. The protocols for the microbead-based multiplex PCR were initially optimized both in bulk solutions and on microdroplets using 9947A female and 9948 male genomic DNA. Sufficient STR fragments were easily produced to enable direct CE analysis after secondary PCR from single beads. Using the optimized procedures, complete STR profiles were obtained from individual GM09947 (female) and GM09948 (male) human lymphoid cells.

Based on this success, single-cell identification studies were performed on mixtures of GM09947 and GM09948 cells at different relative concentrations. The cell mixture study exhibited a good linear relationship between the observed and input cell ratios in the range of 0:1 to 10:1, eliminating the complex interpretation of mixed genotypes. STR profiles of single-cells from both pure and mixed cell populations showed that all alleles were correctly called and allelic drop-in/drop-out was not observed. Additionally, the STR profile of target GM09947 cells could be deduced even in the presence of a high-concentration (three copies/droplet) of cell-free 9948 "background DNA."

Single-Cell STR Analysis, STR Mixture Analysis, Microfluidics

A159 Whole Mitochondrial Genome Probe Capture Next Generation Sequencing Method for Analysis of Limited and Degraded DNA Samples

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After attending this presentation, attendees will be informed about a new method for enriching and sequencing the whole mitochondrial DNA genome for analysis of limited, mixed, and degraded samples using next generation sequencing technology.

This presentation will impact the forensic science community by demonstrating a high throughput, next generation sequencing method for the mitochondrial genome that increases the discrimination power over current methods. Further, the new method introduces a mechanical shearing technique for DNA fragmentation and a liquid-based probe capture step that facilitates the processing of highly degraded samples.

Degraded and/or size-limited DNA samples are often encountered in forensic casework. The application of mini Short Tandem Repeat (STR) and bi-allelic Single-Nucleotide Polymorphisms (SNPs) have increased the success of analyzing these samples; however, these methods target nuclear DNA and, although there is a clear advantage in doing so (high discrimination power), degraded samples often cannot be effectively analyzed. The use of mitochondrial DNA (mtDNA) is beneficial in these cases due to its high copy number and small size. Yet, its highly conserved genome lacks discrimination potential.

The application of next generation sequencing technology allows for the analysis of the entire mitochondrial genome, which compensates for limitations of current mtDNA analysis methods. A probe capture 454 next generation sequencing assay was developed for target enrichment and deep sequencing of the whole mitochondrial genome. The liquid phase probe capture step in the developed method was included into the assay design to enrich for mtDNA and to eliminate the need for amplification primers that target the sample DNA. By doing so, the designed assay has the potential to analyze highly degraded samples while bypassing amplification complications. To further improve the assay's capability of processing degraded samples, a mechanical shearing fragmentation method, independent of sample quality, was incorporated into the assay. The goal of this project was to optimize and validate this assay for use on degraded, mixed, and/or size-limited samples.

The library preparation, the first step in the whole mitochondrial DNA sequencing assay, begins with the fragmentation of the sample DNA into short fragments, typically ~400-500bp. Controlled enzymatic digestion is routinely used for this step in genomic studies, but this approach presents difficulties in analyzing limited and degraded samples. For forensic samples, it is important the assay be capable of processing degraded and limited samples in the same way it processes pristine samples. This would allow a crime laboratory to use a single protocol for any sample it analyzes. To achieve this goal, a fragmentation method using mechanical shearing was optimized and implemented. The Covaris® mechanical shearing uses ultrasonic waves that form cavitation bubbles that cut DNA as they implode. This unique Adaptive Focused Acoustics™ technology has an energy focal point inside the sample tube, making the technology accurate, reproducible, and quantity independent. To test if this mechanical

shearing technology was also quality independent, DNA samples naturally or artificially degraded to different levels were treated with Covaris®. The results showed fragments of uniform size regardless of the initial state of degradation of the sample DNA. In addition to fragmentation, the initial PCR of library preparation was modified for optimal amplification of limited samples. This optimized method was then tested for its ability to process forensically relevant samples.

Results will be presented showing successful analysis of limited, mixed, and degraded samples processed using the optimized fragmentation, capture, and next generation sequencing method. Results from a sensitivity study will be presented demonstrating successful fragmentation and capture of limited DNA amounts with 100% mtDNA sequence coverage. The high success observed for even the lowest DNA amount tested (100pg), which is significantly lower than the manufacturer-recommended DNA sample amount of 1µg, suggests that even lower starting DNA amounts may be successfully analyzed. A mixture study resulted in successfully distinguishing the two sequenced profiles in a 5% mixture (the lowest mixture ratio tested). Finally, degraded DNA sample libraries were successfully created showing proof of concept for the optimized mechanical shearing fragmentation method for processing samples independent of quantity and quality. The results suggest the optimized method has high mixture and concentration sensitivity and has the potential to process degraded samples. The optimized whole mitochondrial DNA sequencing assay shows promising results for further validation and testing on degraded DNA samples encountered in forensic casework.

Next Generation Sequencing, Mitochondrial DNA, Limited & Degraded DNA Sample

A160 Next Generation Sequencing of Human Mitochondrial DNA Extracted From Hair Shafts Using a Multiplexed PCR Strategy and Illumina® Nextera® XT

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After attending this presentation, attendees will gain an understanding of how a multiplex Polymerase Chain Reaction (PCR) approach and next generation sequencing methods can be used to sequence whole mitochondrial genomes from challenging sample types such as short hair shaft fragments.

This presentation will impact the forensic science community by providing insight into how next generation sequencing methods are being used to increase the discriminatory power of human mitochondrial DNA analysis by providing high-quality sequence data that extends to regions of the mtGenome that are not routinely analyzed in forensics laboratories.

Forensic scientists are often faced with the challenge of limited or degraded samples, where a nuclear DNA (nDNA) profile may be difficult to obtain. In these instances, mitochondrial DNA (mtDNA) analysis can be particularly useful, as mtDNA is more easily recoverable from challenging sample types such as hair shafts and bone. Traditional methods focus on sequencing of two hypervariable regions (HV1 and HV2) located in the non-coding control region of the mtGenome; however, HV1/HV2 comprise only about 4% of the mtGenome and mtDNA is less discriminatory than nDNA, which limits the current utility of mtDNA analysis. Studies have shown that expanding analyses to the coding region

of the mtGenome can lead to a dramatic reduction in previously unresolved individuals.¹

Thus, the objective of this research was to expand the amount of information gleaned from a limited sample type, such as a short hair shaft fragment, by combining a multiplexed PCR approach with Next Generation Sequencing (NGS) methods. In this study, mtDNA was extracted from two centimeters of hair shaft collected from two donors using an optimized extraction method designed in WCU's forensics laboratory. Extracts were quantified with a human mtDNA-specific qPCR assay and then amplified using a multiplexed PCR strategy consisting of forty-six total primer sets in ten reactions (four- or five-plexes) and covering the entire mtGenome.² The multiplex reactions were designed using previously described primer sets (Applied Biosystems® MitoSeqr™ Kit) with slight modifications for NGS. Primer set combinations were chosen based on a variety of factors including primer melting temperatures, amplicon size, position in the mtGenome, and evaluation of secondary structure formation. Various amplification conditions and enzymes were attempted during method development using positive control DNA and the Roche® FastStart™ High Fidelity PCR System was found to be most effective. Multiplexed reactions were evaluated with the Agilent® 2100 Bioanalyzer for concentration and sizing information. Amplicons were diluted, pooled in various ways, and processed using Illumina® Nextera XT®, which enzymatically fragments amplicons and tags them with Illumina®-specific adaptors and indexes. This library preparation method requires only 1ng of total dsDNA, making it extremely useful in forensic applications. These processed samples were then sequenced on the Illumina® MiSeq™ platform using a 300-cycle, paired-end v2 reagent kit. Sequencing data was evaluated using Illumina® MiSeq™ Reporter and CLC Bio Genomics Workbench software packages and results were compared to reference sequences generated using blood or buccal samples and traditional Sanger sequencing methods. A total of nine mtGenomes were sequenced in one NGS run with an average coverage of 7,000x across all reported positions, and 89.1% of the five gigabases sequenced exhibited a Phred Q-score ≥ 30.

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mtDNA, Next Generation Sequencing, Multiplex Amplification

A161 Co-Amplification of NUMTs in Low DNA Quantity Specimens

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After attending this presentation, attendees will be informed regarding the incidence and management of co-amplified Nuclear Mitochondrial DNA (NUMT) with authentic mitochondrial DNA while producing a database of more than 550 entire mitochondrial genomes via Sanger sequencing.

This presentation will impact the forensic science community by contributing to the ongoing conversation regarding

mitochondrial DNA recovery and data quality. With the increasing sensitivity of mitochondrial genome typing, especially through the emerging use of next generation sequencing chemistries, the potential for co-amplification of mitochondrial pseudogenes will require greater consideration than has been required in the past.

Forensic mitochondrial DNA (mtDNA) testing requires representative high-quality population databases for estimating the rarity of questioned haplotypes. However, currently available forensic reference population data only include information from the mtDNA control region. To address this deficiency, the Armed Forces DNA Identification Laboratory (AFDIL) undertook a National Institute of Justice-funded, large-scale databasing effort to sequence complete mitochondrial genomes (mtGenomes) spanning three U.S. population groups. Amplification of the complete mtGenome was achieved via eight overlapping fragments and each mtGenome was sequenced in 135 reactions, providing redundant and overlapping forward and reverse sequence coverage across the entire molecule.¹ To assure the generation of the highest quality profiles, nearly all pipetting steps were performed robotically and a rigorous data review process was employed.

The samples used for this databasing effort were anonymized blood serum specimens from the Department of Defense Serum Repository.² Only a small amount of cell-free DNA is typically present in blood serum, as the DNA-containing blood components have been removed by centrifugation. Thus, the samples to which the 8-amplicon Polymerase Chain Reaction (PCR) strategy was applied contained DNA that was generally of high quality but present in very low quantity. Though the mtDNA copy number in any given blood serum extract should readily exceed the available nuclear DNA, amplification of NUMT insert remains a possibility when: (1) total DNA quantity is very low; and, (2) NUMTs both exceed the size of the target mtDNA fragment and have high similarity to modern mtDNA in primer binding regions. Careful PCR primer design can reduce, though not eliminate, this risk.

Rare instances in which NUMTs were co-amplified and sequenced with the authentic mtDNA in the course of generating more than 550 entire mtGenome profiles from low DNA quantity specimens will be presented, along with strategies for handling the samples/data. The NUMTs were distinguished from sample contamination by the appearance of many more heteroplasmic positions than would be expected from a mixture of distantly related modern mtDNA haplotypes, and the minor profiles matched a known mitochondrial pseudogene. Characterization of the presentation and frequency of co-amplified NUMTs will be increasingly important as the sensitivity of whole mtGenome typing improves — such as with the implementation of next generation sequencing technologies in forensic laboratories — and these highly sensitive methods are applied to extremely low template samples.

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The opinions and assertions contained herein are solely those of the authors and are not to be construed as official or as views of the U.S. Department of Justice, U.S. Department of Defense, its branches, the U.S. Army Medical Research and Materiel Command, the Armed Forces Medical Examiner System, the Michigan State Police, the Federal Bureau of Investigation, or the U.S. Government.

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Mitochondrial DNA, Sequencing, NUMT

A162 Genetic Polymorphisms of X-STRs in the Egyptian Population With Discussion of X-STRs' Role in Forensic Case Work

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After attending this presentation, attendees will be informed of different chromosome X genetic polymorphisms that have been observed in Egyptians and resulted in allele dropouts and discordant results between some kits. They will learn methods used for solving such problems. Attendees will gain better understanding of chromosome X Short Tandem Repeat (STR) haplogroups and how they could be used, complementary to autosomal and Y-STRs, to solve complex kinship deficiency cases. Furthermore, they will be introduced to the potential of using X-STRs in identifying female traces in male background.

This study will impact the forensic science community by drawing attention to the importance of conducting genetic studies on different populations for which very poor data is available. It will demonstrate the efficiency of X-STRs in solving special forensic cases. Increasing insight to the forensic impact of chromosome X markers in solving kinship deficiency cases and trace evidence examination will encourage forensic experts to use them in solving cases that were otherwise refractory.

Different structural polymorphisms are present in different degrees in various populations. Depending on the site and size of these polymorphisms, they could affect the performance of different DNA typing kits. Conducting genetic studies on different populations is crucial to understanding the magnitude and effect of these polymorphisms. To date, X-STR markers have not yet been studied in Egyptians.

X-STRs bear the potential to efficiently complement analysis of autosomal and Y-STRs in solving complex kinship deficiency cases. This is because males are hemizygous for the X chromosome and fathers pass their X chromosome completely to their daughters. Furthermore, certain X-STR loci express strong linkage disequilibrium and segregate together as stable haplotypes which exhibit much lower frequencies than single STR alleles and are, therefore, more informative in kinship deficiency cases. Hence, many STR loci and haplogroups in different populations should be examined to have a better view of the most informative combination of loci that could be used.

In addition, theoretically, X-STR markers could be more efficient than autosomal STRs in identifying female traces in male "contamination" since female alleles can only be completely included in the male component if the female happens to be homozygous at all loci; however, this theory still needs practical verification.

In this study, a sample of 250 unrelated Egyptian males was examined with the Investigator® Argus X-12 kit. Null alleles

and new microvariant alleles not included in the current genotyping bin sets of the kit were detected. Discordant results at locus DXS10101 were observed between the Investigator® Argus X12 and Mentype® Argus X8 kits. Two samples had allele dropouts with the Investigator® Argus X-12 kit at DXS10101, and when they were re-amplified using the Mentype® Argus X-8 kit, they yielded genotypes of 28.2 and 30.2. New primers were designed for loci DXS10101, DXS10146, and DXS10148 to correct for the allele dropouts observed at these loci with the Investigator® Argus X-12 kit. DNA sequence analysis revealed structural polymorphisms responsible for the allele dropouts at these loci. In locus DXS10101 a (T@C) SNP in a non-core repeat block, upstream from the repeat region was observed. A four bp (TCTT) deletion was detected at point 149,584,442, nine bp downstream from the repeat region in locus DXS10146. In locus DXS10148, a SNP (G@A) at point 9,238,986 within the core repeat region was detected. Additionally, six X-STRs (DXS10161, DXS10159, DXS10162, DXS10163, DXS10164, and DXS10165) located in the centromere region at Xp11.21-Xq11.1 were examined in a single multiplex. Allele and haplotype-frequencies, as well as forensic statistical parameters, of the 18 X-STR loci tested indicated that they would be highly informative in different forensic applications in Egyptians.

Case reports of kinship deficiency cases will be presented where the use of X chromosome STRs (ChrX-STRs) complementary to autosomal and Y-STR markers was crucial. The efficiency of ChrX-STRs in detecting female traces in male background will be discussed based on testing where female/male DNA mixtures were prepared using DNA cell lines XX28 (Qiagen®) and 2800M (Promega®) with a total DNA concentration of 1ng and the following ratios: 1:1; 1:2; 1:4; 1:10; and 1:19. The percentage of the female minor profile was calculated for each sample. Complete minor profiles were called for all the tested ratios up to 50pg of female DNA to 950pg of male DNA (1:19 ratio).

X-Chromosome STRs, Allele Dropout, Egyptian Population

A163 Single Cell Characterization of *Bacillus* Spores: Novel Forensic Signatures for Biothreat Agents

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After attending this presentation, attendees will understand the effect laboratory preparation methods have on the phenotypic characteristics of *Bacillus* spores, the forensic issues associated with microbial evidence characterization, and the use of Atomic Force Microscopy (AFM) for the characterization of the surfaces of single bacterial spores.

This presentation will impact the forensic science community by introducing a new signature system for determining forensically relevant aspects of a single organism's culturing environment. Since an organism's phenotype can be influenced by laboratory production processes (i.e., culturing conditions and purification processes), knowledge of these processes may provide investigative leads and/or exclude suspects during a forensic investigation.

Phenotypic profiling of bacterial threat agents can be a valuable tool for investigating bioterrorism. By analyzing the non-

genetic features of an organism grown in a laboratory, it is possible to reconstruct key aspects of culturing medium and/or purification process. Recently, several forensic phenotypic signatures have been developed, but most of these methods require a significant number of bacterial cells (i.e., $10^6 - 10^{10}$) which may be unlikely in microbial evidence. New signature systems are needed for the detection and characterization of samples with trace quantities of cells recovered during a forensic investigation.

AFM has emerged as a versatile tool to visualize biological molecules ranging from single proteins and DNA molecules to whole cells, with sub-nanometer resolution. In the context of forensic analysis, the AFM can provide rapid, high-resolution, 3D imaging of cellular samples at the single cell level under a variety of environmental conditions. Additionally, AFM has been used to differentiate between bacterial species and strains; however, the effect of various culturing conditions and purification processes has yet to be fully investigated.

The purpose of this research was to use AFM to identify forensically relevant features of the cell surface that are created during culturing in the laboratory. More specifically, the morphology of the cell surface of *Bacillus cereus* spores was investigated as an indicator of the composition of the growth medium and the purity of the cell culture. The research focused on two strains, T and 14579, of *B. cereus* in four culture media: G medium (G); Peptone G medium (PGM); Tryptone G medium (TGM); and Brain Heart Infusion G medium (BGM). Four purification steps were also studied: zero; one; three; and five washes in nanopure water. For each culture preparation, surfaces of individual cells were analyzed at the micron to nanometer scale. As a result, the spore surface roughness (RMS) was measured.

The results from the effects of culturing medium on the cell surface showed that the roughness of the *Bacillus* spores were consistent between the different growth conditions. Although medium components have been shown not to affect the RMS, results revealed that RMS is affected by purification processes. As the number of washes increased, the topography of the spore changed. Without any washes (i.e., crude purification), both strains of *B. cereus* had several observable small features (<100nm) on the surface and the spores' natural shape was not visible due to the adherence of molecules. However, after one wash, there were fewer features and the spores were ellipsoid in shape. As the number of washes increased, the roughness decreased as a result of the features being removed. Overall, *B. cereus* showed an average RMS value of 15.49nm for zero washes and 11.36nm for five washes. Comparing crude (zero washes) to pure (five washes) spore preparations, the RMS values were significantly different ($P=0.0012$) and could represent a statistically robust signature for differentiating these two types of cells.

Overall, the results of this research suggest that the surface of single spores is affected by the extent of purification and that AFM can be a tool for processing microbial evidence. Since forensic evidence often contains trace quantities of material, the use of AFM in detecting phenotypic differences in *Bacillus* spores from batched cultures treated with various laboratory processes can be a potential technique for the attribution of microbial bio-crimes.

***Bacillus Cereus*, Atomic Force Microscopy (AFM), Microbial Forensics**

A164 The Buffer Zone

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The goal of this presentation is to introduce attendees to

the forensic significance of shotgun shell buffer material used by different ammunition manufacturers.

This presentation will impact the forensic science community by showing the discriminating potential of this type of physical evidence if present at a shooting scene.

The multi-projectile shotgun shell is more complex than standard handgun- or rifle sealed-metallic cartridge ammunition. Besides the enclosure, primer, and propellant, the unfired shotgun shell contains a payload that may consist of a single projectile or metallic shot in a variety of sizes. Metallic shot is predominantly made of lead alloys; however, non-lead alloys or metals (e.g., tungsten or bismuth), with specific properties that influence the behavior of the fired pattern, are also common. Many shotgun ammunition manufacturers use a soft additive (buffer) in their product. Buffer materials usually consist of small polymer granules or spheres. These are commonly added to larger-sized shot (e.g., buckshot) and some smaller loads (e.g., turkey shot). The commingling of the softer polymer buffer with the harder metal pellets provides a cushioning effect that reduces deformation of the pellet's spherical shape. This is important because maintaining the uniformity of the pellets during their sudden acceleration and energetic travel down the barrel will allow the pellets to maintain stable, consistent, and predictable aerodynamic properties during flight to the target.¹ When one of these buffered shotgun shells is fired, the numerous polymer granules are also discharged along with the pellets and wading, but being so much lighter and smaller than the pellets, the buffer does not travel as far.² Nevertheless, in spite of their light weight, buffer granules can become imbedded in close-range wounds or clothing as well as become scattered several yards downrange of the muzzle.³ This research was undertaken to determine whether it is possible to differentiate shotgun shell brands and manufacturers on the basis of the physical and chemical properties of their buffer materials. The ability to associate a sample of buffer, recovered from a crime scene, to a specific ammunition manufacturer may provide valuable information to shooting scene reconstructions. For this research, several brands of ammunition were obtained. The buffer material (both pre- and post-discharge) for each brand was characterized using micrometry, micro-melting point, and micro-Fourier Transform Infrared (FTIR) spectroscopy. The comparison of pre- and post-discharge samples revealed that there were no significant changes in the size or shape after firing. Stereomicroscopy revealed morphological differences among the unfired buffer material used by some of the different manufacturers, as did particle micrometry. Infrared (IR) spectroscopy revealed that, within the analyzed samples, Winchester® used polyethylene for their buffer while Remington® and Federal® used polypropylene. Detailed inspection of the IR spectra revealed differences that differentiated Remington® and Federal® brands.

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Buffer, Shotgun, Reconstruction

A165 Automatic Identification of Bullet Signatures Based on Consecutive Matching Striae (CMS) Criteria

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The goal of this presentation is to introduce a basic model and procedure to process the bullet topography and count the consecutively matching striae in an automated identification system

This presentation will impact the forensic scientific community by showing how this research makes the computerized database search system capable of completely implementing the identification task rather than simply giving a priority order for further manual operations.

Although the forensic science specialty of firearm and tool mark identification in criminal investigation has more than a century of history, it has been challenged over the years for its subjectivity and its difficulty to articulate how identification is made. In 1997, Biasotti and Murdock published their quantitative criteria for identification as expressed in terms of Consecutively Matching Striae (CMS). Since the CMS criteria were established by empirical studies using manual operations and observation, determining the maximum number of well-defined matching striae in a large statistical sample of Known Non-Matches (KNM) was a practical impossibility. Automated systems are expected to go a long way toward establishing objective quantitative criteria and a statistical foundation for identification since they can quickly perform a large number of comparisons. The CMS criteria can then be validated or revised accordingly. These automated measurements, which are more objective, can therefore be used to increase confidence in the validity of the CMS approach originally developed and practiced using direct manual comparisons.

With previous experience in automated bullet identification research, National Institute of Standards and Technology (NIST) tries to address the questions about statistics and subjectivity and to provide a method for automation of CMS as the similarity metric. A computerized identification method based on 3D topography measurement and CMS criteria was developed. The position and shape information of the striae on the bullet land is represented by a feature profile, which is used for determining the CMS number automatically. CMS values are automatically calculated from surface topography images for a set of unknown bullets compared to a set of knowns. The set of test bullets were fired from ten consecutively rifled 9mm pistol barrels. The known matching set consists of 10 pairs of bullets and 60 matching land comparisons. Using the developed algorithm, all 10 pairs of bullets fired from the same gun barrel were successfully identified. Then the model was applied to the 15 "unknown" bullets. Before these experiments, the operator did not obtain any barrel source information about these bullets and, therefore, this phase of the testing was "blind." Each of these 15 unknown bullets should match the two bullets in the control set that were fired from the same barrel. Out of a total of 30 matching pairs, 29 were correctly identified. These tests for the known matching and known non-matching bullets demonstrated the validity of the model. It makes the computerized database search system capable of completely implementing the identification task rather than simply giving a priority order for further manual operations. It increases the objectivity of firearm identification examination. Also, the model lays solid groundwork for future statistical analysis. Even though the database used in this study was limited and was obtained from a specified type of

firearm, it is practical to extend the method to a large database.

Bullet, Identification, Consecutive Matching Striae

A166 The Relevance of Firing Pin and Breechface Scores in the Interpretation of Firearms Evidence

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After attending this presentation, attendees will understand how firing pin and breechface scores can be used to interpret firearms evidence and provide a clearer understanding of variability within firearms.

This presentation will impact the forensic science community by providing greater understanding of the underlying features of impression evidence and how to apply such knowledge during examination.

The terms class- and individual-characteristics are used on a daily basis in the practice of forensic science. There are, however, various interpretations and usages of these terms in the general literature. Generally, a classification process is considered to be grouping of results based upon the outcome of a particular analytical method. The examination of impression evidence is a basic classification problem (match/non-match). Identification, or individualization, is sometimes thought to be the summation of a variety of multiple characteristics until a unitary class is developed. According to the Association of Firearm and Tool Mark Examiners (AFTE) in their *theory of identification*, the concept of *sufficient agreement* is achieved when the agreement: (1) exceeds the best agreement demonstrated between tool marks known to have been produced by different tools; and, (2) the agreement is consistent with tool marks known to have been produced by the same tool.

Conceptually, an analyst needs to understand both the variability of the characteristic being evaluated within the sample under consideration as well as the variability of the same characteristics within the relevant population of similar objects. In order to apply the mechanism of *sufficient agreement* when considering firearms, one needs to evaluate these characteristics. To consider firing pin impressions on a cartridge case, it is necessary to study its variability within a particular firearm. It is possible or likely that factors such as the type of ammunition, primer type, rate of fire, and the structure of a particular cartridge will influence this variability. Second, it is necessary to understand the variability within the particular make and model of the firearm. Third, how the characteristics of the particular type of firearm and the general class of caliber will influence the comparison must be understood.

The challenge in a firearms examination is that, in many cases, there is but a single cartridge case from the scene. The firearms examiner may collect two, three, or five cartridge cases from a suspect firearm to serve as controls for the comparison. The inter-variability of these cartridge cases is evaluated and a representative cartridge case is used to affect the comparison. Commonly, the firearms examiner will evaluate the evidence using a comparison microscope. Another tool in their arsenal is the Integrated Ballistic Identification System/National Integrated Ballistic Identification Network (IBIS/NIBIN) System (Forensic Technologies Inc./Bureau of Alcohol, Tobacco, Firearms and Explosives). The IBIS system provides scores between the firing pin, breech face, and ejector marks of a cartridge against those in the database. Generally, the system is used to identify links between cases.

In this study, 100 cartridges were fired through 16 9mm

firearms. All of these cartridge cases were entered into the system and their breech face and firing pin images were collected. Within the database all unidentified firearms/cartridge cases were designated unknown. All firearms entered were newly purchased. The images were then correlated against the database and the full list of candidates and their scores were obtained. A match was designated between the sample and database cartridge if both were fired from the same firearm (irrespective of the score). All other records were designated as a non-match. The SigSauer® P250 was run against the database and generated 17,478 records. All records were classified as either match or non-match. The resultant plot of the firing pin against the breech face scores indicates the variability in both. A distinct separation between the P250 and all other firearms are given. Unknown plots of background data are used in this study.

Generally speaking, the data suggest that discrimination within the dataset is affected by the size of the database and the type of firearm. The variability in cartridge cases (as demonstrated by their scores) for a single firearm is significant. This variability may mask the effect of other variables.

Firearms Examination, Evidence Interpretation, IBIS

A167 Tests Using the Congruent Matching Cell (CMC) Method for Optical Image Correlations of Cartridge Cases Fired From Consecutively Manufactured Pistol Slides

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After attending this presentation, the attendees will be introduced to the method for the objective quantitation of impressed tool mark comparisons using Congruent Matching Cells (CMC). The results of the comparisons of fired cartridge case breechface tool mark images from consecutively manufactured pistol slides will be reported.

This presentation will impact the forensic science community by the introduction of an accurate and objective mathematical method for the measurement of the similarity of impressed tool marks, in this case fired cartridge case breechface tool marks. This method can be used with comparison microscope photographs.

Outline of Objectives: The National Institute of Standards and Technology (NIST) Ballistics Identification System (NBIS) is developed based on 3D topography measurements and correlation cells. The NBIS aims to provide objective, high-accuracy, and high-speed ballistics identifications. The National Ballistics Evidence Search Engine (NBESE) is proposed by NIST for ballistics evidence searches using the Congruent Matching Cells (CMC) method with system interoperability and error rate report. Feasibility of applying correlation cells for 3D topography correlation is successfully demonstrated by validation tests using 40 cartridge cases fired with 10 consecutively manufactured slides. The results show a significant separation between the Known-Matching (KM) and Known-Non-Matching (KNM) distribution. However, most existing ballistic identification systems are based on intensity image comparisons and a huge number of intensity images are stored in the national ballistic database. It is necessary

to conduct a validation test that demonstrates the feasibility of the CMC method for the identifications of optical intensity images. One primary benefit of an independent, objective, mathematical measurement method is that it can use images captured during the microscopic comparison of firearm or tool mark evidence.

Brief Methodology: A Leica® comparison microscope is used to capture the breech face intensity images of the same set of 40 cartridge cases fired from handguns with ten consecutively manufactured pistol slides. To confirm the equipment accuracy, repeatability and reproducibility tests are performed. In the correlation tests using the CMC method, a total of 780 correlations including 63 KM and 717 KNM were implemented. The effects of different lighting conditions for the image correlations were also tested and analyzed.

Summary of Results: In the initial tests using the single correlation parameter of the whole image, the cross-correlation function maximum CCF_{max}, the KM and KNM distributions cannot be separated. However, by using the CMC method with three identification parameters (the correlation function maximum CCF_{max}, the registration angle θ , and registration position in x , y), the KM and KNM distributions show clear separations with no overlap. There is no false positive or false negative identification in all 780 correlations.

General Conclusions: The results show that the CMC method works well for correlation of both the 3D topographies and optical intensity images. Significant improvement can be achieved by applying the CMC method with three identification parameters on intensity image data compared with the correlation of whole image using a single correlation parameter CCF_{max}. The CMC method also shows a good robustness to lighting condition variance. The identification accuracy can be further improved by optimization of the cell numbers and the thresholds of the correlation parameters.

CMC, Identification, Impressed Tool Mark

A168 Reference Bullets and Cartridge Cases for Automated Ballistics Inspection Systems

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The goals of this presentation are to describe the physical properties of the National Institute of Standards and Technology (NIST) Standard Reference Materials (SRMs) 2460 and 2461 and how they may be acquired and to outline the procedure for using the SRMs to test optical identification systems in crime laboratories

This presentation will impact the forensic science community by showing how SRMs 2460 and 2461 will help crime laboratories to: (1) verify that automated optical equipment for bullet and cartridge case image acquisition and correlation is operating properly; (2) establish ballistics measurement traceability; and, (3) achieve accreditation.

NIST SRMs 2460 and 2461 are physical standards that provide replica tool markings of fired bullets and a fired cartridge

case, respectively. SRM 2460, the standard bullets, were certified and first distributed in 2006. SRM 2461, the standard cartridge cases, were certified in July 2012.

Each unit of SRM 2460 is a copper pellet in the shape of a 9mm bullet, which contains six Land Engraved Areas (LEAs) fabricated using an ultra-precision, numerically controlled diamond turning machine. The machined profile of each LEA is a replica of a profile measured on a fired bullet. Six different profiles were used from six different firearms, three from the Bureau of Alcohol, Tobacco, Firearms and Explosives (ATF) and three from the Federal Bureau of Investigation (FBI). Because of the highly-repeatable manufacturing process, all units of SRM 2460 are essentially identical.

Each unit of SRM 2461 consists of a circular electroformed nickel plate, replicated from the head of a fired master cartridge case, which contains the surface topography of a breech face impression, a firing pin impression, and an ejector mark. The electroformed plate is cemented to a brass cylinder holder so that the assembly resembles a real fired cartridge case. The master cartridge case was fired at the National Laboratory Center of ATF. The SRM cartridge cases produced from that master by electroforming have virtually the same surface topography signatures. The surface topography of each SRM unit produced was measured at NIST to confirm the similarity of the tool marks.

The key property of the SRMs is the similarity of the tool marks on different SRM units, enabling comparisons and quality control of measurement results obtained at different sites. In a controlled system, images of different units taken by the same model inspection instruments, in different places, at different times, and by different operators should be highly similar. This similarity can be easily quantified if the different inspection tools share common databases, such as the National Integrated Ballistics Information Network (NIBIN) developed and coordinated by the ATF in collaboration with local crime laboratories. NIBIN includes approximately 200 ballistic inspection systems linked together into regional databases and having common software for acquisition and correlation testing. The NIBIN incorporates two models of ballistics inspection instruments. Individual laboratories can assess whether their identification process is in control by comparing SRM measurement results with the control limits developed under a project known as the National Ballistics Imaging Comparison.

The SRMs may also be used to test systems that directly measure surface topography of cartridge cases and bullets. Topography images acquired on such systems can be compared with reference images obtained at NIST and available on the website, pml.nist.gov/srm2461. For SRM 2461, two parameters are used to quantify the similarity of the cartridge case surface topography images: the maximum value of the areal cross correlation function, $ACCF_{max}$, and the relative signature difference D_s . When two correlated cartridge case signatures are exactly the same (point by point), D_s must be equal to 0 and $ACCF_{max}$ must be equal to 100%.

The surface topography of all SRM 2461 units and three reference units were measured with a confocal microscope at NIST. The topography images were processed using band pass filtering to minimize noise, form and waviness, thus emphasizing the fine roughness features of the tool mark topographies. The SRM unit topography images were then correlated with those of the three reference units. For the breech face, firing pin, and ejector mark regions of the distributed SRM units, a lower limit for $ACCF_{max}$ and an upper limit for D_s , each with a 95% confidence level are reported in the SRM certificates. Similar statistics are reported for SRM 2460.

Standards, Topography, Ballistics

A169 2D/3D Topography Comparisons of Tool Marks Generated by Ten Consecutively Manufactured Chisels and Drift Punches

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The goal of this presentation is to evaluate whether a mathematically objective metric, the maximum value (CCF_{max}) of the Cross Correlation Function, can be used to identify a tool that generated a striated or impressed tool mark. The metric will be applied to the measured surface topography of tool marks generated under laboratory conditions on pristine surfaces. The study addresses the two types of tool marks and tools that produce them: chisels for the production of striated tool marks, and punches for the production of impressed tool marks. The study produced tool marks in a randomized fashion, and the results were tabulated blindly using an automated data analysis and identification system.

This presentation will impact the forensic science community by providing validation to the science of tool mark identification through the use of an objective identification method and criterion. Using these statistical methods, key recommendations in the 2009 National Academy of Sciences Report are addressed.¹

A tool mark is created when plastic deformation and/or displacement is caused in a soft material by a tool. Tool marks generally appear in two forms: striated and impressed. Striated tool marks are formed when a tool-working surface is placed on another surface and moved parallel to that surface. Impressed tool marks are formed when the tool surface is forced perpendicular into another surface.² Consecutively manufactured tools have the most likely chance of producing similar tool marks, which have the greatest possibility of causing false-positive identifications. Ten consecutively manufactured chisels and punches were obtained from Western Forge (a supplier of Craftsman® Tools). The tools were separated into striated tool marks generated by the chisels and impressed tool marks generated by the punches. For each of the 10½" (12.7mm) chisels, a set of two known tool marks were created on a polished copper plate through a controlled dragging motion of the chisel produced by a motorized jig. After the identities of the chisels were randomized and hidden, an additional set of 20 unknown tool marks (two marks per chisel) were created. For each tool mark, a 2D stylus probe instrument was used to measure the surface topography along a line orthogonal to the striations. A total of 1,600 profile comparisons were performed in a 40 x 40 matrix. For each comparison, the two profiles were automatically registered to obtain the maximum value (CCF_{max}) of the Cross Correlation Function. The CCF_{max} values for the known match and known non-match comparisons were used to establish test statistics for the reliable identification of the unknown tool marks based on CCF_{max} values.

For each of the ten punches, a set of two known tool marks were created on a polished copper plate through a controlled drop of the punch, which was mounted on a linear rail. After the identities of the punches were randomized and hidden, a set of 20 unknown tool marks (two marks per punch) were created. A disk-scanning confocal microscope was used to measure the 3D topography of the tool marks. The data were then trimmed to remove any uninformative areas. Before the comparisons, each measurement was automatically pre-processed. Dropouts and outliers, which typically occur in areas of low reflectivity or high slope variations, were identified and masked. A total of 1,600 surface comparisons were performed in a 40 x 40 matrix. For each comparison, the pre-processed data sets were automatically registered in position

and orientation to obtain the maximum value ($ACCF_{max}$) of the Areal Cross Correlation Function. The known match and known non-match $ACCF_{max}$ values were used to establish test statistics for the reliable identification of the unknown tool marks based on $ACCF_{max}$ values.

In a blind study of ten consecutively manufactured chisels and punches, the maximum value (CCF_{max}) of the Cross Correlation Function was successfully applied to link the measured surface topography of a tool mark to the tool that created it. All of the unknown striated chisel tool marks and impressed punch tool marks were correctly identified back to the tool that created them. These results provide an objective mathematical validation of the science for both striated and impressed tool mark comparisons.

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Tool Marks, Cross Correlation Function, Topography

A170 An Improved Vacuum Casting Method for the Replication of Reference Bullets

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After attending this presentation, attendees will understand the application of vacuum casting to produce high-quality polymer replications of bullets, materials used, process steps and improvements, and comparisons of microscopy images and topography measurements.

This presentation will impact the forensic science community by providing a tool for forensic examiners to replicate ballistic evidence, such as tool marks generated by a firearm on a bullet or cartridge case. There are many instances where the transfer of ballistic evidence from one laboratory to another would be beneficial, e.g., in solving "cold cases," obtaining another assessment, or when there are suspected links in violent crimes.

The National Institute of Standards and Technology (NIST) developed an improved vacuum-casting replication method based on earlier work at the Bundeskriminalamt in Germany.¹ The replication process will be used to replenish the supply of NIST Standard Reference Material (SRM) 2460 bullets. The key property of the SRM bullets is the similarity of the striated markings on each bullet, enabling comparisons and quality control of measurement results obtained at different sites. The reference bullets were originally manufactured using a highly-repeatable, but expensive, diamond turning process. Extensive testing showed that polymer replication through vacuum casting is a viable and cost-effective alternative. The NIST Standard bullets are also well defined, and therefore allowed the validation of the polymer replication process through quantitative measurements and statistical methods.

The two-step process through which a replica polymer bullet is made is described. A negative mold of the bullet is first made using silicone, after which a positive replica bullet is made

using polyurethane. The specific vacuum-casting techniques discussed are critical in forming clean replica bullets, free of micro-bubbles and contamination. Both qualitative and quantitative comparisons of the replicas with the original bullets are presented. The comparisons are based on conventional optical bright field microscopy images and measurements of the surface topography using confocal microscopy and surface profilometry.

Durability is a key requirement of reference samples. Since polyurethane plastics are not as hard as metal bullets, there is the potential for degradation of the replicas due to aging, environmental factors (temperature, humidity), abrasion, and contaminants. The results of several controlled experiments to address these concerns are presented.

High-quality replication enables sharing of ballistic information when the actual transfer of the physical evidence is not possible, either due to logistics, chain of custody issues, or simply because the evidence is too valuable to ship. Replicates can make measurements possible that cannot be performed on the original samples. Replication also enables the dissemination of virtually identical reference samples or proficiency testing sets to improve quality control and training.

Reference:

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Bullet Replication, Silicone Molding, Polyurethane Casting

A171 Attenuated Total Reflectance-FTIR Spectroscopy for Gunshot Residue Analysis: Potential for Ammunition Determination

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The goal of this presentation is to describe the development of a novel and alternative method for Gunshot Residue (GSR) analysis.¹ After attending this presentation, attendees will have a better understanding of the application of Attenuated Total Reflectance-Fourier Transform Infrared Spectroscopy (ATR-FTIR) for GSR analysis, identification, and discrimination. The implementation of advanced statistics to differentiate experimental spectra collected from non-equivalent GSR samples will be discussed.

This presentation will impact the forensic science community by potentially greatly impacting the accuracy and effectiveness of shooting incident investigations.

The successful application of Raman micro-spectroscopy combined with advanced statistics for the analysis and differentiation of GSR particles originating from two different calibers (0.38 inch and 9mm) of ammunition was reported last year.² For that research, only light-colored (tan) GSR particles were selected for analysis, due to strong fluorescence interferences resulting from the analysis of darker-colored GSR particles. The conclusion was that the technique has the potential to provide a strong chemical and statistical link between GSR samples and specific firearm-ammunition combinations, as well as the ability to exclude a specific firearm-ammunition combination from generating the unknown GSR sample. Reported Raman spectra were found to have contributions from both organic and inorganic components of GSR; however, the method did not rely upon the detection of these heavy metals, illustrating its potential as a novel alternative

to current GSR analysis techniques.

This is a report on the application of ATR-FTIR spectroscopy combined with advanced statistics for GSR analysis. Evidentiary analysis by FTIR is fast, user-friendly, relatively inexpensive, and non-destructive. Little sample preparation or sample quantity is required, and the resulting FTIR spectra offer a molecular fingerprint for the compound in question. Additionally, the technique offers a combination of unique features such as portability, database-search functionality, and multivariate analysis capabilities. Applications of FTIR to forensic chemistry include identification of paint, explosives, smokeless gunpowder propellant, and illicit drugs. Previous investigations of GSR by FTIR were focused on qualitative chemical composition and shooting distance determinations. In the present study, FTIR spectra were collected from *individual* GSR particles originating from three different firearm-ammunition combinations. Preliminary results indicate that the new technique is more comprehensive than all currently accepted and innovative methods of GSR identification. Successful differentiation of FTIR spectra from three firearm-ammunition combinations (0.38 inch, 0.40 inch, and 9mm calibers) was achieved. Informative FTIR spectra could be collected from *all particles* regardless of their color, fluorescence contribution, and morphology, representing significant improvements relative to the reported earlier Raman-based approach.² In this work, informative FTIR spectra were collected from previously unanalyzable firearm discharge samples, consisting of exclusively dark particles (0.40 inch). Principal Component Analysis (PCA) revealed grouping, by data collected from the same caliber. Projection to Latent Structures Discriminant Analysis (PLS-DA), also known as Partial Least Squares Discriminant Analysis, was able to differentiate GSR data from the three firearm-ammunition combinations (calibers) at a rate of 100% for the internal validation and 93.3% for the external validation. These results are supported by Leave-One-Out (LOO) cross validation.

This emerging technique illustrates the possibility for an on-scene, non-destructive, identification and chemical characterization method for GSR. This method has the potential to greatly impact the forensic science community by increasing the accuracy (and discriminatory power) of GSR detection. The most direct application for this research is a method to exclude a specific firearm-ammunition combination as producing an evidentiary GSR sample. The comparison of a laboratory-generated GSR sample discharge and an evidentiary GSR sample can be made without extensive preliminary studies.

References:

1. Bueno, J.; Sikirzhytski, V. and Lednev, I.K. Attenuated Total Reflectance-FT-IR Spectroscopy for Gunshot Residue Analysis: Potential for Ammunition Determination. *Analyt Chem*, 2013. Published on Web. DOI: 10.1021/ac4011843.
2. Bueno, J.; Sikirzhytski, V. and Lednev, I.K. Raman Spectroscopic Analysis of Gunshot Residue Offering Great Potential for Caliber Differentiation. *Analyt Chem*, 2012, 84, 4334-4339. (Made a journal cover, C&E News, and Canada Discovery Channel — Daily Planet program).

Gunshot Residue, Trace Evidence, Vibrational Spectroscopy

A172 Background Gunshot Residue — A Statistical Analysis

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After attending this presentation, attendees will have learned about the levels of gunshot residue in a population of detainees who have been arrested on non-firearms-related

charges.

This presentation will impact the forensic science community by illustrating how the use of gunshot residue evidence can be enhanced with the statistical analysis.

Gunshot Residue (GSR) analysis is a forensic sub-discipline in Trace Evidence. The forensic utility of GSR evidence is to provide information as to whether there is a primary association of a person or surface with the discharge of a weapon or subsequent handling of a discharged weapon. There are many factors that can affect the persistence of GSR on a given surface which include, but are not limited to, time between the event and sampling, activity (cleaning), and environment (weather). Additionally, challenges may be raised suggesting that peace officers, who carry and use firearms, and their places of employment, can be a source of contamination.

GSR examiners concur with certain premises (e.g., that residues arising from the detonation of lead styphnate, barium nitrate, and antimony sulfide primers, PbBaSb, are characteristic of GSR and that the presence of GSR does not indicate that a person has fired a weapon), but there is no consensus on background levels of GSR. At the 2005 Federal Bureau of Investigation (FBI) Laboratory's Gunshot Residue Symposium, the attendees reported minimum thresholds ranging from one to fifteen PbBaSb particles, with a majority stating that a single GSR particle is a sufficient threshold.¹ The National Academy of Sciences Report of 2009, *Strengthening Forensic Science in the United States: A Path Forward*, did not directly address GSR evidence, but the report recommends that forensic reporting be standardized and uniform.² The wide range of reporting thresholds leads to confusion in the interpretation of GSR results.

Thresholds can be utilized to determine statistical significance of a result when compared to a population of interest. If the average number of PbBaSb particles on the hands of members of the general population were known, then the Poisson probability distribution function

$$P_n = \frac{\mu^n}{n!} e^{-\mu}$$

μ is the average number of PbBaSb particles

n is an experimentally determined number of particles

provides this statistical inference. Since the entire population cannot be evaluated, a suitable sample is needed to estimate it.

Efforts to determine the frequency presence of GSR particles in the population without recent, direct exposure to a shooting event have been reported in two studies.^{3,4} The prevalence of GSR in samples from "non-shooting" populations have focused upon police officers that have not fired their weapons for lengthy periods of time (up to 90 days). A total of four PbBaSb particles were found on the hands of 124 police officers in the combined studies.

A third study focused on surfaces commonly encountered by suspects in police custody, e.g., rear seats of patrol cars, interview rooms, lock-ups, etc.⁵ In this study, a total of 201 surfaces were analyzed and a total of 56 PbBaSb particles were found. There is a clear difference in the statistical inferences that can be made from this study when compared with the data from the hands of police officers who had not recently fired weapons. A fourth unpublished study of subjects appearing before magistrates after being in police custody yielded no PbBaSb particles on the dominant hands of 100 individuals. The widely varying results from these studies were the starting point for this work.

Hands of 175 detainees being processed into the Harris County Inmate Processing Center (IPC) were sampled. Consent was obtained and then samples were collected in the pre-trial services area of the IPC. The samples were analyzed by Scanning Electron Microscopy/Energy Dispersive X-Ray spectroscopy (SEM/

EDX). No characteristic GSR particles were found on any of the samples.

These results indicate that the statistics require the use of inequalities. A combined average of the results of this study and those found in references three, four, and six will be used: $\mu=0.0126$. The Poisson distribution for the mean leads us to infer that 98.4% of the "uninvolved" population would be expected to have no characteristic GSR particles. Given a suggested threshold of three particles for primary association, the odds of finding this number of characteristic particles on an uninvolved person are on the order of 1:3,000,000.

This project was supported by NIJ Award #2010-DN-BX-K222 awarded by the National Institute of Justice, Office of Justice Programs, U.S. Department of Justice. The opinions, findings, and conclusions or recommendations expressed in this publication/program/exhibition are those of the author(s) and do not necessarily reflect those of the Department of Justice.

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Gunshot Residue, Statistics, Arrestees

A173 Discharge of a Pistol Out of a Car Window With the Breach Within the Interior of the Car: Analysis of Gunshot Residue on a Car's Interior Surfaces

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The goal of this presentation is to show attendees where to sample for Gunshot Residue (GSR) in the interior of a car to document a firearm discharge within the vehicle.

This presentation will impact the forensic science community by providing locations for the sampling of a car interior to document GSR contamination from a firearm discharge while its breach is within the car.

The defendant, the driver of the questioned car, allegedly extended his right arm over the passenger seat and fired a single round from a .380 pistol out the passenger window. One witness, however, claimed that the defendant got out of his car and fired the pistol. A casing was not found at the scene. The attorney for the defendant requested an analysis for GSR of the alleged car, a 1997 Honda Civic®, used in the shooting. The brand of the .380 pistol was not known. A bullet was not recovered. A CCI® .380 Blazer® standard-primed cartridge with a full copper jacket bullet was hypothesized as being used in the shooting because the defendant had a box of this ammunition in the trunk of his car when arrested two weeks after the shooting. The CCI® Blazer® ammunition is distinctive for U.S.-manufactured ammunitions in that it has aluminum casings with the Berdan primer design.

Sampling for GSR of the defendant's car: The alleged car used in the shooting, a 1997 red Honda Civic®, was sampled at a police car storage yard. Thirty dabs each were made with a GSR sampler of the right door frame and the headliner over the

passenger door. A heavy contamination of road dust prevented other surfaces from being sampled.

Considering the possible sources of the small number of GSR-like particles found in the defendant's car, it cannot be determined these few characteristic and consistent GSR particles were deposited on these surfaces by a discharge of a firearm from within the interior of the car or were transferred by hand contact.

Test of a similar car: A 2005 Honda® Civic® of similar design to the questioned car was driven to a rural road and a Bryco® Arms Model 48 .380 pistol fired one time by a person in the driver's seat out the passenger window. The breech of the pistol was within the car's interior which simulates the shooting scenario in the defendant's car. As with the defendant's car, there were 30 dabs per sample.

The single discharge of the pistol in the 2005 Honda® Civic® contaminated the interior surfaces of the car with GSR. The passenger window headliner and frame received many GSR particles. However, the dashboard sampling of a vehicle will provide more reliable evidence a firearm was discharged within a vehicle than other surfaces in the vehicle. A dashboard burden of GSR can only be from airborne deposition. Other surfaces where contact by hand is possible cannot be discounted for transfer of GSR from this source.

Gunshot Residue, Car Interior, Firearm Discharge

A174 Transmission Electron Microscopy Characterization of Gunshot Residues From Brazilian Ammunition

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After attending this presentation, attendees will gain a better understanding of both the structure and the formation process of Gunshot Residue (GSR) and also be provided with new perspectives for GSR analysis.

This presentation will impact the forensic community by questioning previous knowledge about GSR and discussing some of the implications of the new findings.

GSR originates from the rapid condensation of elements from the primer mixture vaporized during firearm discharge, and are usually described as spheroidal with diameters ranging from 0.5 to 10.0µm. Scanning Electron Microscope (SEM) coupled with Energy Dispersive X-Ray Spectrometry (EDS) has been recognized as the most useful method for its identification. Transmission Electron Microscope (TEM) is not a usual forensic tool because it requires sample preparation and interpretation is not straightforward. Nevertheless, it combines high magnification capability and the ability to explore the internal structure together with the chemical composition, a combination which is essential for nanometric analysis. The goal was to analyze GSR from different types and calibers of ammunition produced in Brazil and sold worldwide employing TEM to characterize elemental composition and morphology of nanometric residues. Characteristic GSR, combining spherical morphology and three-component content, were found in all nine samples analyzed by SEM but only in two when analyzed by TEM. By SEM, characteristic and consistent particles were found to be in average $\geq 4\mu\text{m}$. TEM showed particles much smaller, approximately 100 to 200nm, probably due to the collection technique and the fact that larger particles may be lost

from the grids by gravity. A typical characteristic GSR nanoparticle observed by TEM is spherical and heterogeneous in contrast. The nanoparticles appear to be composed of even smaller agglomerates. There is an accepted notion that these particles are amorphous. Results show that this reasoning is due to a limitation of SEM's resolution. Selected Area (Electron) Diffraction (SAED) pattern shows that GSR are essentially crystalline, opposed to the ASTM E-1588 standard description. The presence of diffuse rings was seen in almost all particles observed, indicating that crystalline nanoparticles are present in most GSR. Although only slightly apparent in conventional TEM images, the presence of smaller nanoparticles agglomerates forming larger GSR is clearly visible in Dark Field (DF) images. Employing High-Angle Annular Dark-Field Scanning Transmission Electron Microscopy (HAADF STEM) mode, characteristic three-element GSR composed of 5-10nm nanoparticles was observed and also seen in the DF image taken from diffraction spots. Inconsistent particles composed of PbSb, the DF image from a diffraction spot with interplanar distance of 0.29nm showed that they are also composed of agglomerates sized approximately 2-10nm. This characteristic was observed in nearly all particles observed regardless of the ammunition caliber used, suggesting that this may comprise a specificity derived from the formation process of GSR. Most GSR automatic search systems are limited by the minimum particle size of 0.5-0.7µm detected. The results show that particles of 100-200nm are present in a concentration equivalent to microscopic GSR, and are essentially agglomerates of much smaller nanoparticles of approximately 2-10nm. The interplanar distances in the Diffraction Patterns (DP) taken from several particles in all samples were calculated. Some of the values found were a good match to phases that could form the GSR, considering that the reflections with the highest intensities were found in the DPs of the GSRs. The phases considered were lead oxides, lead and antimony oxides, barium oxides, metallic lead, and metallic antimony. The results demonstrate that there is a large population of GSR nanoparticles overlooked. These nanoparticles are prone to stay airborne and can have the potential to coalesce in larger particles and contaminate the scene and non-related bystanders. Most of the crime labs filter the $<1\mu\text{m}$ particles in order to avoid airborne contamination and also to reduce errors due to the resolution limitations of the SEM. Nevertheless, these nanoparticles, mostly composed of Pb, may also constitute another problem unrelated to forensic investigation. The threshold limit value of exposure for lead in a workplace is 50mg/m³. The method for collecting particles consists of a 220nm passing filter. Although TEM analysis is much more complex than SEM, it could also be used as a tool to investigate evidence collected from gun-related suspects and crime scenes as it can provide unique information from different ammunition.

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TEM, Gunshot Residue, Nanoparticle

A175 Counterfeit Drugs and Their Impact on Forensic Science

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After attending this presentation, attendees will learn about counterfeit drugs and the challenges these drugs present to the global forensic science system. In addition, attendees will learn the best analytical approach to detect, identify, and source counterfeit drugs in the field.

This presentation will impact the forensic science

community by describing complex factors and providing the background necessary to enable the forensic science community to successfully address the counterfeit drug problem.

The counterfeit drug problem is complex and integrates matters of science, law, criminal justice, public health, and public policy. There is a need to delineate the integral aspects of the counterfeit drug trade if an effective solution is to be proposed.

The counterfeit-drug problem is extremely complex and addressing this problem requires an integrative approach. Forensic science, local laws and law enforcement, drug-development regulations, international law, public treaties and policies, funding of drug development and Intellectual Property (IP) rights, and enforcement all need to be considered if an appropriate solution to this problem is to be presented. This presentation will describe each of these factors and provide the background necessary to enable the forensic science community to successfully address this problem.

Counterfeit drugs present unique and complex challenges to the forensic scientist. The impact of these goods is devastating, ranging from localized fatalities resulting from small-scale product adulteration to large-scale public health crises due to the development of drug resistance and fatal diseases caused by substandard medications. These goods present a significant regional and global problem and are a serious threat to public health and safety. They are forcing forensic science, criminal justice, and law enforcement organizations to reevaluate current policies and practices so that successful investigation and adjudication in these types of cases is possible. The nature of these goods as well as the internet and other factors that have streamlined global trade are rendering current practices ineffective.

The field of forensic science struggles to establish analytical methods to identify composition and provenance of counterfeit goods. No method has been shown to be universally applied to achieve this goal. The use of field-portable instruments to detect and identify counterfeits in the field is important emerging technology. These instruments must work effectively and must meet the standards for admissibility of evidence in court when field-test results are presented as scientific evidence.

Results of research performed on counterfeits to identify and source these drugs in the field as well as methods based upon infrared spectroscopy, Raman spectroscopy, and gas chromatography-mass spectrometry will be presented. Method limitations as well as discrimination potential will be reviewed. In addition, challenges to admissibility in court will be discussed.

Counterfeit Drugs, Forensic Science, Public Health

A176 Analysis of Tobacco Exposure in Human Hair Using Total Vaporization-Solid Phase Microextraction (TV-SPME) Gas Chromatography/Mass Spectrometry (GC/MS)

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After attending this presentation, attendees will understand a new sampling technique for the analysis of volatile components using Total Vaporization-Solid Phase Microextraction (TV-SPME). Specifically, this presentation will describe the analysis of tobacco exposure in human hair by TV-SPME.

This presentation will impact the forensic science community by describing, in detail, the theory behind TV-SPME and its applicability to volatile analytes in artifacts of forensic interest.

SPME is a sampling technique in which volatile components are absorbed onto a fiber and then subsequently desorbed into an analytical instrument such as a liquid or gas chromatograph. The thermodynamics of SPME are largely dependent on analyte partitioning between the liquid sample, the headspace above the sample, and the SPME fiber. A new technique has been proposed whereby a liquid sample is vaporized. By completely vaporizing the sample, the partitioning between the sample and the headspace is eliminated, thus simplifying the thermodynamic equilibrium. This presentation will discuss the theory and application of TV-SPME. Additionally, there are many parameters that must be optimized in developing a SPME method, including SPME fiber chemistry, incubation temperature, incubation time, extraction time, desorption time, and sample volume. Using a statistical experimental design is the best way to determine the optimal parameters without performing every possible parameter combination, or a "vary one parameter at a time" method. A statistical experimental design also allows for interactions between parameters to be realized; for example, how sample volume and incubation temperature interact with one another. A response surface methodology experimental design was used to optimize the parameters of the TV-SPME technique.

Current methods of determining tobacco exposure from hair require large sample sizes and extensive extraction procedures. The methods being developed require only 10mg of hair and the extraction is conducted by digesting the hair in sodium hydroxide followed by liquid-liquid extraction with chloroform. Nicotine and its metabolite (cotinine) are quantitated using TV-SPME coupled to GC/MS. Typical protocols for analyzing tobacco exposure from human hair use liquid injections with GC or LC. In GC, injection volumes are usually limited to several microliters. This can be problematic, especially when sample size is limited, because the concentration of nicotine and cotinine in hair is very low (<50ng/mg hair). Using total vaporization SPME, lower analyte concentrations can be easily detected because the sample volume is much larger (>100µL). This greatly increases the analyte amount injected onto a GC column, thereby making total vaporization SPME a very sensitive technique. Tobacco exposure will be determined by comparing nicotine and cotinine concentrations from a hair digest, and classifying people as tobacco users, non-users, and ex-tobacco users. In addition, it would be beneficial to classify tobacco users as smokers and smokeless tobacco users. This has been accomplished using urine, but there are no reports using hair.

Solid Phase Microextraction, Human Hair, Tobacco Exposure

A177 High Throughput Analysis of Street-Quality Drug Mixtures by DART® GSX Analysis and MassWorks™ Post Acquisition Characterization

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After attending this presentation, attendees will have an understanding of the analytical capabilities of the Direct Analysis in Real Time (DART®) GSX™ instrumental platform from IonSense® for the rapid analysis of street-quality drug mixtures. The inherent, high throughput capabilities of this technique will be compared to current Gas Chromatography/Mass Spectrometry (GC/MS)

analytical methods.

This presentation will impact the forensic science community by demonstrating the qualitative high throughput analysis of street-quality controlled substances by DART® ionization which allows for the rapid characterization of unknown evidence samples by decreasing laboratory time and costs of instrumental operation compared to current GC/MS platforms.

The new DART® GSX™ platform allows for the ambient ionization of solid and liquid samples. Ionized species are then detected and analyzed by an Agilent® Gas Chromatography/Mass Selective Detector (GC/MSD) quadrupole mass analyzer. Protonation of molecules for mass analysis is achieved by the formation of metastable helium species which then react with atmospheric water and create protonated water clusters. These protonated water clusters interact with the analytes of interest, creating protonated species, which enter the mass analyzer and are separated from neutrals through the use of ion optics. Simulated street quality drug mixtures were created in increasing complexity and concentration with the addition of common adulterants and diluents in order to mimic unknown evidence samples commonly encountered in the analysis of controlled substances. Common adulterants and diluents including, but not limited to, lidocaine, benzocaine, procaine, levamisole, caffeine, creatinine, mannitol, inositol, and boric acid were utilized in the creation of street-quality cocaine mixtures. A continually developing mass spectral library for common adulterants and diluents by DART® ionization was created in order to aid in the post-acquisition analysis of cocaine mixtures. Further confirmation of mass spectral data created by the DART® GSX™ platform was performed on an equivalent DART® ionization source interfaced to a Thermo LCQ™ Classic with an ion trap mass analyzer.

Post-acquisition processing of mass spectral data for cocaine mixtures were performed through Agilent® ChemStation® analysis software. The stepwise subtraction of suspected adulterants/diluents contained within cocaine mixtures was performed to enable rapid and simple mass differentiation and analyte confirmation of cocaine. Further analysis by Selective Ion Monitoring (SIM) enabled the successful identification of cocaine and its fragments within samples where the concentration of adulterants and diluents present were greater than 500 times the concentration of the amount of cocaine present within the same sample. Several sampling methods and substrate types, such as stainless steel wire mesh, cotton swabs, polyester swabs, and glass capillaries, were examined to enable rapid automated sample introduction.

MassWorks™ post-acquisition analysis software from Cerno Bioscience was used for accurate mass assignment and chemical formula determination. Using known calibration standards along with the MSIntegrity calibration technology, Calibrated Line-shape Isotope Profile Search (CLIPS) was performed to increase spectral accuracy on the unit resolution GSX quadrupole system and to identify molecular formulas for known mixtures.

Comparative analysis of the DART® GSX™ instrumental platform versus current GC/MS platforms demonstrated the characterization of complex cocaine mixtures in a fraction of the time required by equivalent GC/MS methods. With a decrease in the use of consumables, time spent on sample preparation/introduction, and cost of operation, DART® ionization eliminates several drawbacks of current GC/MS methodologies for the characterization of controlled substances in forensic laboratories.

DART®, Cocaine, High Throughput

A178 Multivariate Statistical Procedures for the Classification of Controlled Substances in Simulated Street Samples

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After attending this presentation, attendees will be more familiar with the application of statistical procedures for the identification and classification of controlled substances.

This presentation will impact the forensic science community by further demonstrating the potential of statistical procedures for the evaluation of forensic evidence. While the focus will be on controlled substance data collected using infrared spectroscopy, the statistical procedures demonstrated are applicable to a wider range of evidence types and analytical techniques. Recommendations will be discussed for the application of such procedures in the analysis of spectral data.

As with many other forensic disciplines, the identification of controlled substances in submitted samples often involves comparison of analytical data to known reference standards analyzed under the same conditions. For the analysis of controlled substances, Infrared (IR) spectroscopy is among the analytical techniques considered to have the highest discriminating power (SWGDRUG Category A technique). However, as street samples often contain numerous other components, comparison to spectra of reference standards can be challenging without prior extraction and isolation of the substance.

In this research, the potential of multivariate statistical procedures for the identification of controlled substances in street samples analyzed by IR spectroscopy was investigated. First, a set of controlled substance reference standards was analyzed by IR spectroscopy with an attenuated total reflectance sampling accessory. Selected reference standards were also mixed with caffeine to generate a set of simulated street samples that were analyzed in a similar manner. All spectra were smoothed and normalized prior to further statistical analysis, which involved both exploratory and classification procedures.

Exploratory data analysis procedures are unsupervised in nature as no *a priori* knowledge of the data under investigation is necessary. Instead, natural groupings of the data are identified, which can be beneficial as the first step in the analysis of complex data. In this research, Hierarchical Cluster Analysis (HCA) and Principal Components Analysis (PCA) were investigated as the exploratory procedures. HCA assesses similarity among samples, displaying results as a dendrogram from which the similarity level at which such sample groups form can be determined. In contrast, PCA identifies sources of variance in the data set to reduce dimensionality without losing discriminatory information. Results are displayed in the form of a scatter plot in which chemically similar samples are positioned closely and distinctly from chemically different samples.

While exploratory procedures have potential and utility in a forensic setting, the ability to classify samples with statistical confidence is also desirable. To that end, two classification procedures, *k*-Nearest Neighbors (*k*-NN) and Soft Independent Modeling of Class Analogy (SIMCA) were also investigated in this research. In *k*-NN, new samples are considered members of the group to which the nearest *k* neighbors belong. For SIMCA, previously defined groups within the data set are firstly modeled using PCA. New samples are introduced to each model and the significance of the fit is evaluated statistically, with samples considered members of the groups to which there is the highest probability of membership. This highlights one major difference between *k*-NN and SIMCA: the former is a hard classification procedure, meaning that every sample will be classified to one group. In contrast, SIMCA is a soft classification procedure, meaning that the classification is not forced. As such, new samples may be classified into one group, more than one group, or no

groups at all.

This presentation will demonstrate application of each exploratory and classification procedure to the spectral data set to investigate association and subsequent classification of the simulated samples to the appropriate reference standard. Results of each procedure will be discussed and compared, highlighting the importance of appropriate data pretreatment and representative statistical models for classification. Distinct advantages and disadvantages of the procedures for this application will also be presented, along with recommendations to facilitate future implementation in forensic laboratories.

Multivariate Statistics, Controlled Substances, IR Spectroscopy

A179 Rapid Controlled Substance Identification by X-Ray Diffraction

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After attending this presentation, attendees will understand some principles of the analytical technique of X-Ray Diffraction (XRD) and the necessary elements of data collection as well as proper data treatment and analysis for the identification of controlled substances by XRD.

This presentation will impact the forensic science community by demonstrating a recently accepted method for the identification and potential rapid quantification of controlled substances by means of a non-destructive technique, without requiring a need for a controlled substance license to identify drugs, which will aid in monitoring clandestine laboratory activity as well as drug trafficking.

The definition of controlled substances may vary across region and country boundaries, but generally includes narcotics, narcotic-precursors, steroids, and designated clandestine laboratory reagents. As controlled substances evolve (e.g., designer drugs), they often complicate analytical determination. For example, clandestine substances often contain both organic and inorganic materials and very often the materials of interest can have similar molecular weights and structures. These complications present a strain on standard analytical-chromatographic techniques. Most confirmatory techniques are destructive with extensive sample preparation times and often result in the loss of forensic evidence and are subjective to cross-contamination. For similar reasons, the international organization Scientific Working Group for the Analysis of Seized Drugs (SWGDRUG) recently recommended XRD as a primary technique for controlled substance identification.

Advantages of the XRD technique include significantly reduced sample preparation, limited or no risk of cross contamination, and rapid throughput. Furthermore, as a non-destructive technique, it is possible to use additional phase and structural information to gather extensive material intelligence aiding in the identification trafficking activities. Since phase identification is basically a comparison between the unknown measurements and known (reference) diffraction patterns, a reference database is mandatory for phase identification. There are several commercial and non-commercial alternatives available, but in case of "new" active pharmaceutical active ingredients, reference patterns typically cannot be found in commercial databases. A user would have to measure the reference patterns of the different polymorphs for the creation of a user-made reference database or use single crystal data of the compounds of interest. The creation of an XRD database dedicated to controlled substances will be discussed. This database contains more than 550 substances that are

directly related to forensic criminal drug analysis. The bulk of this presentation represents an abbreviated summary of work carried out in partnership with the Drug Analysis Services Branch of Health Canada and focuses on narcotic analysis using modern XRD instrumentation equipped with advanced optics.

X-Ray Diffraction, Drugs, Forensics

A180 2014 Update From the Scientific Working Group for the Analysis of Seized Drugs (SWGDRUG)

Sandra E. Rodriguez-Cruz, PhD, Drug Enforcement Admin, Southwest Laboratory, 2815 Scott Street, Vista, CA 92081*

After attending this presentation, attendees will be aware of up-to-date information regarding SWGDRUG and its documents and recommendations.

This presentation will impact the forensic science community by discussing revisions to SWGDRUG recommendations, new supplemental documents, and other work products in development, as they relate to the analysis of seized drugs.

The Scientific Working Group for the Analysis of Seized Drugs (SWGDRUG) was formed in 1997 as a joint effort between the U.S. Drug Enforcement Administration (DEA) Office of Forensic Sciences and the Office of National Drug Control Policy (ONDCP). The mission of SWGDRUG is to recommend minimum standards for the forensic examination of seized drugs and to seek their international acceptance.

Current SWGDRUG recommendations are available to the general public via the group's website (www.swgdrug.org). The SWGDRUG core committee has revised its recommendations pertaining to the use of reference materials. Laboratories throughout the world are encountering difficulties obtaining and verifying new reference materials. These tasks have become more overwhelming with the appearance of new designer drugs like synthetic cannabinoids and bath salts. Revisions to the recommendations include minimum procedures for the verification of these materials, as well as alternative guidance when newly emerging materials are encountered and verification is limited to structural elucidation procedures.

During 2013, the SWGDRUG core committee finalized, approved, and posted Supplemental Document SD-4. This document provides three examples of measurement uncertainty calculations for purity determinations.

The SWGDRUG mass spectral library continues to be regularly updated and currently contains more than 1,880 spectra. Included in the library are many of the recently encountered synthetic cannabinoids, substituted cathinones, and hallucinogenic phenethylamines. Laboratory analysts throughout the world can download this library from the SWGDRUG website and into their laboratory instruments. Feedback from analysts and library users continues to be highly positive. The library will continue to be updated on a regular basis and contributions from the forensic community are strongly encouraged.

Since November 2012 and continuing throughout 2013, SWGDRUG has made available numerous drug monographs containing detailed information and analytical data for reference materials which have been analyzed, verified, and authenticated by the Drug Enforcement Administration Special Testing and Research Laboratory. These monographs, available via the SWGDRUG website, are intended to be used for the verification of acquired reference materials.

With the goal of assessing the current controlled substance analogue issue, SWGDRUG has drafted a document containing general recommendations regarding analogues and structural class

determinations. This document was posted for public comment during the spring of 2013. The document emphasizes the need for analysts to fully understand how analogues and structural classes are legally defined in their jurisdiction before rendering opinions about the classification of a substance.

During 2013, SWGDRUG also made available multiple training documents to be used as resources during the development of laboratory training programs. The documents include questions covering general as well as specific subject matters relevant to the analysis of seized drugs. Subjects included are color tests, infrared spectroscopy, gas chromatography/mass spectrometry, and separation techniques, among many others.

The SWGDRUG core committee is comprised of representatives from federal, state, and local law enforcement agencies in the United States, Canada, Brazil, Great Britain, Germany, Austria, Switzerland, Australia, and Singapore. The following forensic organizations are represented: the European Network of Forensic Science Institutes (ENFSI), the Academia Iberoamericana de Criminalística y Estudios Forenses (AICEF), the Asian Forensic Science Network (AFSN), and the United Nations Office on Drugs and Crime (UNODC). Core committee members also include forensic science educators and representatives from forensic science organizations across the United States, the American Society of Crime Laboratory Directors (ASCLD), the American Society for Testing and Materials (ASTM), and the National Institute of Standards and Technology (NIST).

Criminalistics, SWGDRUG, Drug Analysis

A181 Incorporating Solid Phase GC-IR Into a Controlled Substance Analysis Scheme

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After attending this presentation, attendees will understand the value of incorporating solid phase Gas Chromatography-Infrared Spectrometry (GC-IR) into their analytical drug scheme for the identification of substances which may not be readily identifiable by traditionally employed forensic methods.

This presentation will impact the forensic science community by detailing the advantages of using a combinational approach that includes solid phase GC-IR to assist in the analysis of designer drugs. The forensic drug community is experiencing an unprecedented growth in the submission of designer or synthetically manufactured substances, many of which require analytical tools not commonly employed to assure an unequivocal identification of the submitted evidence.

The nature of routine controlled substance analysis is slowly changing in the midst of an ever-increasing submission to the forensic laboratory of synthetically prepared substances. These substances may contain only slight modifications to a parent structure as "manufacturers" try to avoid existing laws which regulate a particular compound. These modifications may be difficult to detect or confirm as the isomers of closely related compounds are often indistinguishable by Gas Chromatography/Mass Spectrometry (GC/MS), the routine analysis method of choice for many forensic laboratories.

One analytical approach to identify these substances applies multiple techniques to achieve an unequivocal identification. The use of solid phase GC-IR as a complementary tool to routine GC/MS can provide an additional foundation for substance identification. The ability to separate mixtures via a GC coupled with the identification power of solid phase IR, makes GC-

IR a valuable addition to the procedures already in place in the laboratory. IR spectra obtained via this technique can be compared to existing solid phase libraries and are of high resolution to detect small changes in compound structures.

The discriminatory power of a method that employs both GC/MS and solid phase GC-IR will be demonstrated through the examination of a number of designer drugs. The presentation will show the solid phase transmission spectra obtained from the GC-IR analysis of synthetic cannabinoid agonists, cathinones, and phenethylamines, detailing the spectral differences that allow for differentiation of these compounds. The analysis of the cannabinoid receptor agonist JWH-018 and related isomers will be discussed using this dual instrumental approach. Positional isomers of fluoromethcathinone will be compared and contrasted. In addition, the structure and spectra of phenethylamines such as phentermine and methamphetamine will be reviewed. Spectra will be detailed from these substances which will demonstrate the capacity of the GC-IR technique to provide valuable information to allow examiners to identify substances that are not easily differentiated by GC/MS analysis alone. From these comparisons, the presentation will show how infrared spectra from solid phase GC-IR has sufficient resolution to differentiate the positional isomers, diastereomers, and analogs of these compounds.

Comparisons between other IR techniques, Attenuated Total Reflectance (ATR), and gas phase GC-IR will also be discussed and the advantages and disadvantages of each detailed. The role of this technique in routine analysis will also be reviewed showing a possible approach to maximize information and minimize additional examiner work.

Controlled Substance Analysis, Solid Phase GC-IR, Designer Drugs

A182 Identification and Quantification of Synthetic Cannabinoids in Herbal Products With NMR

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After attending this presentation, attendees will understand the fundamental principles of Nuclear Magnetic Resonance (NMR) spectroscopy, specifically their applications for rapid detection of synthetic cannabinoids in herbal incenses. The attendee will also appreciate the power of combining proton NMR with 2D NMR techniques to promptly elucidate the accurate structures of cannabinoids with no ambiguity and quantify cannabinoids in herbal incenses through the use of proton NMR.

This presentation will impact the forensic science community by introducing a simplified NMR spectroscopic approach to quickly identify and quantify synthetic cannabinoids and isomers found in herbal incenses.

In addition to the five classes of cannabinoids scheduled in S. 3187, three new compounds were temporarily added to the Schedule I controlled substance list in 2013. Newer compounds are being synthesized promptly and relentlessly to circumvent the ban, which exacerbates the analytical difficulties for forensic labs, some of which are still experiencing backlogs.

This hypothesis implements the use of proton-NMR and

proton-proton correlation NMR spectroscopy (COSY) to positively identify and quantify synthetic cannabinoids in herbal products with minimal sample preparation, all of which can be completed within two hours with eight-minute or shorter NMR scans.

Prior to the NMR study, a simple extraction was employed in 1-mL NMR solvent such as deuterated chloroform (CDCl_3) or d_6 -acetone to remove synthetic cannabinoids from 50mg of solid herbs. After a one-minute proton-NMR scan in CDCl_3 , synthetic components were detected and processed. Subsequently, signature chemical shift values were retrieved and used to identify the synthetic cannabinoids. Most signature peaks from indole cannabinoids were found to be in the 6.5-8.5ppm or 3.8-4.5ppm chemical shift ranges, which do not overlap with the herbal background signals from blank herbal extracts. An eight-minute COSY scan in CDCl_3 on the same sample further confirmed the identities of the cannabinoids, particularly in the 2-D "fingerprint" regions. After four-minute proton-NMR scans, as many as three cannabinoids in herbal products can be quantified with the presence of maleic acid as an internal standard and d_6 -acetone as the NMR solvent.

With the combination of proton- and COSY NMR, 38 herbal samples were rapidly screened for synthetic cannabinoids. Synthetic cannabinoids were found in all but three herbal incenses. Among the herbal samples investigated, AM-2201, JWH-122, JWH-210, RCS-4, XLR-11, and UR-144 were found to be the predominant ingredients. The quantitative NMR results are comparable to chromatographic quantification results, both yielding 0.5-40mg of cannabinoids per gram of herbal product. The standard deviation varies due to the uneven spreading of synthetic components on herbs during the manufacturing process.

In conclusion, accurate identification and quantification of synthetic cannabinoids in powder and herbal samples can be rapidly achieved with proton and COSY NMR. This simple NMR method can differentiate isomers such as JWH-019, JWH-180, and JWH-122. As an alternative to conventional GC/MS methods, this NMR method can help forensic chemists achieve accurate identification and quantification within two hours which increases the analytical throughput and could potentially help to decrease sample backlogs.

Cannabinoids, NMR, COSY

A183 Profiling of Methylamphetamine in Australia Using Stable Isotope Ratio Mass Spectroscopy

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The goal of this presentation is to present data and conclusions which can assist forensic drug laboratories in their ability to classify the precursor synthetic route of methylamphetamine samples. The presentation will also give the attendees an Australian perspective on the profiling of methylamphetamine samples and the trends seen in samples seized at the Australian border and domestically.

This presentation will impact the forensic science community by providing the skills to best interpret stable isotope ratio values of methylamphetamine with regards to obtaining strategic intelligence. On a broader scale, this presentation will

display the potential of using stable isotope ratio values in not only strategic intelligence gathering but also in tactical comparison investigations.

This presentation will discuss the use of stable Isotope Ratio Mass Spectroscopy (IRMS) as a tool for profiling methylamphetamine samples at the National Measurement Institute Australia (NMI). The NMI carries out chemical profiling of many illicit drugs with the goal of providing strategic and tactical intelligence to the Australian Federal Police (AFP). Such information can assist law enforcement agencies in controlling the diversion of legitimate chemicals for illegitimate uses.

At the NMI, methylamphetamine profiling involves examining the data from several chemical signatures. These include organic impurity profiling, chiral analysis, elemental analysis, and identification of any adulterants or diluents. IRMS is another signature employed in the profiling of methylamphetamine samples. The technique has proven to be most valuable in providing strategic intelligence for samples where conventional chemical profiling has failed to provide any useful information in assisting in the classification of methylamphetamine synthetic route or precursor used. By carefully measuring the stable isotope ratio values of methylamphetamine we are able, in most cases, to determine the precursor as well as its synthetic origin in the case of ephedrine/pseudoephedrine. The isotope ratio values are also of great value in tactical comparisons, especially in cases where little to no information has been obtained from the other chemical profiling signatures.

The major synthetic pathways used in the clandestine manufacture of methylamphetamine employ either ephedrine/pseudoephedrine or phenyl-2-propanone (P2P) as their precursors. Ephedrine/pseudoephedrine is an industrial chemical that is produced via three main processes: (1) naturally extracted from the *Ephedra* plant; (2) a semi-synthetic procedure involving the fermentation of sugars in the presence of benzaldehyde; and, (3) a fully synthetic procedure made from 1-phenylpropanone. This presentation will bring together the results and conclusions drawn from previous and current research work conducted at the NMI.¹⁻⁵ It will demonstrate how classifications of the precursor ephedrine/pseudoephedrine and its synthetic origin are made by carefully measuring the stable isotope ratios of C, H, and N in methylamphetamine. Isotopic data for methylamphetamine samples known to have been produced from phenyl-2-propanone made from phenylacetic acid and alpha-phenylacetoacetonitrile will also be presented.

The presentation will stress the importance of marrying all profiling results when drawing conclusions regarding the classification of the synthetic route of methylamphetamine samples and their precursor synthetic origin. It will give examples of cases where erroneous conclusions can be made from profiling information when not considered holistically. Finally, trends of methylamphetamine synthesis and precursor usage encountered at the Australian border will be presented.

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Drug Profiling, IRMS, Methamphetamine

A184 Analytical Investigations of Synthetic Cathinone Street Drugs

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After attending this presentation, attendees will learn of the complexities in the analysis of cathinones, or bath salts, and how initial true discovery methodology was refined into two different targeted approaches for the most common street materials utilizing GC and HPLC separations as well as a direct solids analysis-time of flight detection. This presentation will convey the difficulties in the method development, and how ultimately two different approaches were successful for final determination.

This presentation will impact the forensic science community by showing how conventional drug-testing protocols are not appropriate for the analysis of the cathinones due to their increased reactivity, and also due to many of the possible compounds not existing as reference standards. The approaches utilized in this work are based on discovery approaches in order to determine compound identity; this is then transferred to instrumentation to allow for final identification and quantification.

Various outlets across the globe are carrying products sold as bath salts, plant food, and jewelry cleaner. These products contain synthetic drugs, which have become prevalent in the United States since 2009 when they came from Europe.¹ The drug names (i.e., bath salts) illustrate the variety of marketing tactics that manufacturers employ to lure customers. Products exhibit colorful packaging labels and every bag states the product is "Not for Human Consumption" to allow for legal possession and consumption by circumventing potential control mechanisms.² Not only have these substances been sold in head shops, but they are also retailed in gas stations, adult stores, independently owned convenience stores, and online retailers as of late.³ The internet has provided a significant means of circulating new compounds quickly and effectively. In March 2009, there were reportedly fewer than ten online vendors; by June, that number had grown to dozens, with new sites opening every week.⁴

The products are mainly composed of synthetic cathinones. The cathinone backbone can be functionalized in four different places to create hundreds of possible structures and numerous compounds have already been identified in the products.⁵ Three prominent compounds were temporarily categorized as Schedule I drugs by the U.S. Drug Enforcement Administration (DEA) on October 21, 2011. The banned drugs include MDPV, mephedrone, and methylone.⁶ The products are sold as tablets, capsules, and powders, and modes of abuse include ingestion, inhalation, injection, and insufflation.⁷ They have also been sold by independent dealers in combination with other illicit controlled substances such as ecstasy in tablets and capsules. In 2011,

hundreds of calls per month were made to poison control centers across the country and there have been reported deaths in some cases, mostly due to conjugation of the drugs with other agents such as cocaine, alcohol, and MDMA.^{8,9} Numerous hospitalizations have also been reported.¹⁰ Only recently has pharmacological information come to light on the newly scheduled drugs.

Analysis of such designer drugs and the identification of individual compounds may help ban their production and abuse, but the analysis can be quite challenging. Identification of these drugs can help increase knowledge on their pharmacology to provide better treatment options and possibly decrease the number of calls to poison control centers across the country. In regard to the forensic community, crime labs already have a large workload. An efficient methodology will benefit analysts by increasing laboratory throughput.

This presentation will discuss the development of the extraction of the drug compounds from various commercial media, followed by separation using both Gas Chromatography with Mass Spectrometric detection (GC/MS) and Liquid Chromatography with Time-Of-Flight Mass Spectrometry (LC/TOF/MS). The developed chromatographic method provides qualitative and quantitative analysis of synthetic compounds in the samples based on the use of appropriate standards. A preparatory High-Performance Liquid Chromatography (HPLC) method for the fractionation of multi-component samples and the use of direct infusion MS/MS in further identification of unknown samples will also be discussed.

In addition, the presentation will also discuss the use of direct sample analysis, coupled to TOF/MS as another technique for rapid determination of street drug composition. Utilizing accurate mass TOF/MS, it is possible to characterize the street samples without sample preparation, resulting in a very rapid analysis.

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Cathinones, GC/MS, DSA-TOF-MS

A185 Infrared Identification of Heroin Salts

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The goal of this presentation is to examine the infrared identification of the salts of heroin, which can be problematic due to the formation of an amorphous state in the course of analysis. The precipitation behavior of heroin salts is fundamental to this discussion and the related issues of crystallization and hydration will be addressed. The discriminating features of infrared spectra will also be examined for the different halide salts of heroin.

This presentation will impact the forensic science community by describing an extraction procedure and recrystallization scheme that can facilitate the infrared identification of the salt forms of heroin in drug evidence.

Infrared spectroscopy has become an invaluable technique that is routinely used in the analysis of controlled substances. The technique can be readily applied to solid-dosage forms, which generally allows the chemical form of the substance (base, salt form, and hydration state) to be determined as part of the drug identity. The infrared spectrum of the solid-dosage form is a function of the crystalline structure of the substance. The molecular arrangement is highly symmetric in the crystal and presents a reproducible infrared spectrum that is usually characterized by a multitude of finely-detailed, sharp spectral features unique to the substance in a specific chemical form.

A limitation to the infrared analysis of solid-dosage forms is in the formation of an amorphous (non-crystalline) state for the drug substance that lacks the symmetry of periodic molecular packing. The spectra of amorphous solids are characterized by broad spectral features that lose much of the fine detail found in the spectrum of the crystalline state, which can make an infrared identification unreliable. Most controlled substances readily condense into a crystalline state and their infrared identification is routine; however, the salt forms of some opiates, and especially heroin, often precipitate as amorphous solids, which complicate their infrared analysis.

The formation of heroin salts is strongly influenced by the phenomenon of hydration, whereby water molecules participate in the molecular packing in the solid state. The common solid-dosage form of heroin is the hydrochloride salt, which forms a crystalline monohydrate phase and presents a distinctive infrared spectrum.¹ Hydration usually occurs during precipitation of a molecular crystal when water is present in the solvent, and thereby available to be integrated into the crystal structure as it assembles. Many drug substances that form a hydrated molecular crystal can also occur as an anhydrous phase (without any water), which consists of a different crystalline structure that is distinguishable by infrared spectroscopy.² Heroin hydrochloride can also condense into an anhydrous solid (its precipitation requires exceptionally dry solvents), although the infrared spectrum lacks significant fine detail that suggests an amorphous material.

A critical factor with the precipitation of the hydrochloride monohydrate crystal is the presence of sufficient water in the solvent system to accommodate the composition of the solid. Another issue is the rate of precipitation, where rapid condensation produces an amorphous hydrate with a spectrum unsuitable for identification.

Impurities can also be a complicating factor, and especially O⁶-monoacetylmorphine, a common breakdown product of heroin that arises from de-acetylation in the presence of moisture.

Recrystallization schemes that minimize impurities and favor the precipitation of the crystalline monohydrate phase will be presented. Heroin evidence often occurs as mixtures with other substances and can be relatively simple or complex (e.g., black tar heroin). One recrystallization procedure has proved generally successful for heroin evidence, although low purity samples of tar heroin (heroin less than 30%) remain problematic.

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Heroin Hydrochloride, Infrared Spectroscopy, Molecular Crystallization

A186 Provenancing Cannabis With Plant Isoscapes in New Zealand

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WITHDRAWN

A187 Analysis of Thirteen Designer Drugs Using GCMS, UV-Vis, and FTIR

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After attending this presentation, attendees will learn how a combination of three techniques (Gas Chromatography/Mass Spectrometry (GC/MS), Ultraviolet-Visible Spectrometry (UV/Vis), and Fourier-Transform Infrared spectroscopy (FTIR)) were used to identify 13 different designer drugs.

This presentation will impact the forensic scientific community by demonstrating that a combination of techniques provides excellent discrimination potential for a challenging set of new designer street drugs. In particular, UV/Vis spectrometry, a technique that is often overlooked as a screening tool, is restored to prominence as a powerful, rapid screening tool that can differentiate between drugs that may not be resolvable by gas chromatography.

A variety of designer drugs, sold with labels such as "bath salts," "research chemicals," and "not for human consumption," have been encountered recently in crime labs in Minnesota. The compounds chosen for this project were synthetic cathinones and analogs to Schedule I controlled substances that have been reported in the literature and mentioned in Erowid.org posts, including methcathinone, 3-fluoromethcathinone, 4-fluoromethcathinone, 4-methyl-methcathinone (mephedrone), butylone, ethylone, buphedrone, n-ethylcathinone, 2-fluoroamphetamine, 4-fluoroamphetamine, 2-fluoromethamphetamine, 4-fluoromethamphetamine, and methoxetamine.

One of the challenges with the new designer drugs is that they often show no color development with traditional color screening test reagents, such as the Marquis test. The 13 drugs analyzed demonstrated absorbance in the UV range, and in some cases the UV spectra provided sufficient discrimination to distinguish drugs with similar structures that were not resolvable by GC.

The UV-Vis sample preparation consisted of using 1mg/mL reference drug standards and diluting to maintain linear response. Each standard was diluted with methanol and sulfuric acid, with the amount varying.

The GC/MS sample preparation consisted of using 0.1 μ L of 1 mg/mL reference drug standard with a septum capped vial. Four methods of analysis were used, with three of the methods having parameters adapted from previous references for the available instrumentation.^{1,2,3} The methods varied in the initial temperatures, temperature ramp rate, final temperature, and hold time, and all used a split injection with a split ratio of 100:1.

The FTIR sample preparation consisted of allowing the 1mg/mL reference drug standards to evaporate overnight and placing the powder residue directly onto a diamond Attenuated Total Reflectance (ATR) stage crystal.

The 13 compounds were distinguishable using the combination of GC/MS, UV/Vis, and FTIR techniques. 3- and 4-fluoromethcathinone were distinguishable using UV-Vis, which showed variation in the number of maximum absorbance peaks. Ethylone and butylone showed distinct mass fragmentation patterns despite their similar structures and molecular weights. Buphedrone and n-ethylcathinone also demonstrated differences in their mass fragmentation patterns. The 2- and 4-fluoroamphetamines were resolvable by their gas chromatography retention times. For 2- and 4-fluoromethamphetamine, significant differences surfaced in the IR absorption wave numbers and the percent transmittance.

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Designer Drugs, Synthetic Cathinones, Spectroscopy

A188 Detection Canine Field Accuracy to Flowers Producing Methyl Benzoate (A Cocaine Odorant)

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The goal of this presentation is to inform attendees of the capabilities of canine detectors, in terms of their field accuracy and selectivity, by discussing their response to common landscaping and decorative plants that contain the active odor signature of an illicit substance.

This presentation will impact the forensic science community by strengthening the weight and validity of evidence discovered by detector canines in a court of law through the exploration of the figures of merit of detector canines, which

includes their accuracy, selectivity, and reliability.

Biological detectors, such as canines, are valuable tools for the rapid identification of illicit substances because they can be trained to reliably detect a wide variety of odors and track them to the source. Previous studies have shown that detection canines do not induce an alert response to the parent compound or illicit substance, but to specific components comprised of the substance's signature odor profile. Such signature odors are Volatile Organic Compounds (VOCs) identified in the headspace of the substance in question. For example, methyl benzoate has been identified as the active signature odor of cocaine.¹ While this revelation has assisted in the understanding of canine detectors, helped to improve their training and efficiency, and led to the production of pseudo-drug training aids, it has also raised many questions. Recently, the accuracy and reliability of canine detectors has been challenged within the forensic community and the legal system.

One recent example of the scrutiny faced by detection canines was referenced in the case *State of Florida vs. Joelis Jardines*.² This case revolved around the argument of whether a warrantless canine drug-detection sniff performed at the front door of a private residence was a breach of the Fourth Amendment. The Respondent's position, believing that this scenario is, in fact, unconstitutional, used scientific canine literature to back up this argument to aid in discrediting the use of canines in this scenario. It was stated that if a canine is alerting to the active odor signature that the canine associates with the contraband, and not the contraband itself, the canine's accuracy and selectivity is under question, especially since many of these compounds have been found in common household products. Specifically, methyl benzoate, the sweet-smelling VOC associated with cocaine, has recently been found to be the most abundant compound produced by popular landscaping flowers, such as snapdragons (*Antirrhinums*), as a means of attracting pollinators.³ Therefore, the question arose whether a canine would falsely alert to a snapdragon flower bed, bouquet, or arrangement if a canine alerts to methyl benzoate, as there was no previous scientific evidence to support this claim.

The purpose of this study was to examine the VOCs released from various types of newly grown snapdragon flowers, primarily methyl benzoate, and assess its potential at eliciting a false alert from specially trained detector canines. An optimized method using Headspace Solid-Phase Microextraction coupled with Gas Chromatography-Mass Spectrometry (HS-SPME-GC/MS) was used to assess the different types and abundances of compounds generated from snapdragons at various stages throughout their life cycle, as well as to determine the day at which the highest amount of VOCs was produced. The findings from this study were used to evaluate the accuracy of the response given by detector canines, especially when the flowers were at their highest methyl benzoate production rates. The results suggest that although methyl benzoate is a heavy contributor to the odor profile of snapdragon flowers, other compounds contribute heavily, if not more, than that of methyl benzoate. A canine's ability to distinguish between unfamiliar pools of odor which slightly resemble the illicit substance, like those produced by snapdragon flowers, as well as the potential of a false alert in a similar field scenario, was examined. More than fifty canine trials were performed to evaluate four different types of snapdragon flowers. Results showed that 0% of the canines alerted to the flowers, while 100% alerted to cocaine, the positive control. Though methyl benzoate was found to be released by the snapdragon flowers, the results revealed that canines do not need further training to differentiate between similar odor pools validating their discrimination power and use in the field.

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Detection Canines, Methyl Benzoate, Illicit Substances

A189 You Got DNA From WHAT?

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After attending this presentation, attendees will have knowledge regarding interesting and challenging samples from which their peers were able to obtain interpretable DNA profiles.

This presentation will impact the forensic science community by demonstrating that their forensic biology peers have great senses of humor.

This inaugural Annual Friday evening *You Got DNA From WHAT?* session, proposed by Criminalistics Section Fellow Daniel Petersen, is intended to be informal, entertaining, and informational (in that order!). If you've ever found yourself at loose ends on the Friday night before the meeting wraps up on Saturday and you've ever made this exclamation, or wish you had, this session is for you!

You Got DNA From WHAT? will allow attendees to gather and pay homage to how amazing DNA is, and to reflect on the fact that DNA has been extracted from SO many novel — and interesting — items. As testing sensitivities improve and technologies continue to change, one constant is the need for intuitive analysts to properly sample items of evidence. This session gives those intuitive DNA dudes and dudettes an opportunity to present brief PowerPoint® slide-based synopses on unique and challenging evidence samples. This event would allow DNA analysts to: boast of their DNA success; discuss challenges in obtaining results from a particular item of evidence; ponder how they might improve on their method in the unlikely event they ever encounter this type of evidence again in their lifetime; and gloat vis-à-vis their analytical superiority.

DNA, Sensitivity, Technology Change

A190 Overview of NIST Activities in the Forensic Sciences

Mark D. Stolorow, MS, MBA, NIST, Law Enforcement Standards Office, 100 Bureau Drive, MS 8102, Gaithersburg, MD 20899-8102*

After attending this presentation, attendees will understand how the National Institute of Standards and Technology (NIST) serves the international forensic science community.

This presentation will impact the forensic science community by informing forensic science practitioners, managers, and other key criminal justice system stakeholders of the myriad of forensic science services provided by NIST and the current activities across the federal government in which NIST collaborates in providing national leadership to the forensic science community.

Among the core strengths of NIST are a long history in forensic science and being the leading institute in measurement science, research, standards development, and technology

development. Since the publication of the National Academy of Sciences (NAS) Report in 2009 entitled, *Strengthening Forensic Science in the United States: A Path Forward*, there has been increasing interest across the country in addressing the report's recommendations for improving the forensic sciences. The Executive and Legislative branches of the federal government together with a large number of professional forensic science organizations are among those participating in the quest for an organized and meaningful path to strengthen the forensic sciences as stressed in the NAS Report.

The Forensic Science Program in the Office of Special Programs at NIST has the responsibility of coordinating the agency's efforts in forensic science research, technology, and standards development. NIST is taking on an increasing role in providing national leadership in the forensic science community through its activities in collaboration with key stakeholders in federal, state, and local government, criminal justice, academia, industry, and the private sector. Attendees will receive a broad review of NIST's current forensic science activities, including: its role in the National Commission on Forensic Science; the organization and administration of the new forensic science Guidance Groups; the manufacture and distribution of Standard Reference Materials (SRMs); Standard Reference Databases; workshops and symposia convened at NIST and via webinars directed at specific forensic science disciplines; biometrics and human identification factors; technical working groups dedicated to human factors and cognitive bias in a variety of forensic science disciplines; Automated Fingerprint Identification System (AFIS) interoperability; biological evidence preservation; crime laboratory design, remodeling and construction; forensic science research projects, publications and presentations at forensic science conferences and symposia; and collaborative inter-laboratory studies involving many of the attendees' laboratories.

It is the goal of NIST to facilitate the objective quantification through measurement science and statistical uncertainty determination for many of the current and evolving forensic science protocols, to conduct research to facilitate understanding the physical and chemical principles underlying the measurement approaches, and to assist in the process of standards development in forensic science.

National Commission, Scientific Guidance Groups, Measurement Science

A191 Strategic Research Directions in Forensic Science at the NIST Information Technology Laboratory

Martin Herman, PhD, 100 Bureau Drive, MS 2000, Gaithersburg, MD 20899*

After attending this presentation, attendees will understand how the National Institute of Standards and Technology (NIST) Information Technology Laboratory's (ITL's) unique mission and expertise in computer science, mathematics, and statistics can be applied to improving forensic science.

This presentation will impact the forensic science community by providing strategic research directions that will result in improving the scientific, measurement, and statistical foundations that underlie forensic methods, standards, practices, and technologies, resulting in greater reliability, accuracy, validity, and throughput of forensic analyses.

The following strategic forensic science research challenges may be addressed by ITL:

- **Scientific Underpinnings** — The fundamental measurement science underpinnings of the pattern-based and digital forensic

sciences must be solidified. Research is needed to address issues of accuracy, reliability, and validity in these disciplines.

- **Statistical Foundations** — There is a need for development of quantifiable measures of uncertainty for forensic measurements and for the conclusions of forensic tests and analyses. Statistical tools and practices need to be formulated and integrated into tests and analyses performed by practitioners.
- **Validation Studies** — Validation studies are needed that determine how well forensic methods, practices, and technologies in pattern-based and digital forensic disciplines, interoperability standards and technologies, and automated computing technologies perform under a variety of conditions of use.
- **Human Bias and Error** — Much must be done to understand the sources of human bias in forensic science procedures and to develop mitigation strategies. Testing methodologies need to be developed to determine and evaluate human error, including the sources of error. Metrology needs to be developed that can be used to characterize and quantify the amounts of human error from the various sources.
- **Computing Technologies and Reference Data** — Computing technologies and reference data sets in forensics can lead to greater accuracy, reliability, functionality, and throughput. New automated computing technologies and data sets relevant to forensics need to be developed, tested, and validated.
- **Usability** — Forensic systems and methods with enhanced usability can lead to greater accuracy, reliability, and throughput, as well as greater satisfaction for forensic practitioners. Studies are needed to provide a better understanding of how to improve forensic usability, and where such improvement can enhance forensic practices. Standards, practices, and technology need to be developed or updated to achieve improved usability.
- **Interoperability** — Enhanced interoperability of forensic systems and methods are potentially capable of providing greater reliability and throughput through improved collaboration and sharing of information. Standards, practices, protocols, and technology need to be developed or updated to achieve improved interoperability.

The forensic science research performed by NIST ITL focuses on four critical themes:

1. Image and Pattern Analysis — Many forensic disciplines are based on image, pattern, or impression evidence, including friction-ridge patterns, face imagery, voice, scars-marks-tattoos, bitemarks, tire marks, shoe prints, tool marks, and handwriting. Image and pattern analysis is also necessary for video and audio analytics, for comparison and identification of software files in digital forensics, and for 2D and 3D forensic shape metrology and analysis.

2. Measurement and Uncertainty — All the forensic science disciplines require a solid underpinning of statistical foundations, measurement science, and measurement uncertainty. This theme covers topics such as measurement errors, error rates, errors due to human bias, errors in forensic methods and technologies, statistics-based tools and practices, and assessing the statistical validity of forensic methods, standards and tools.

3. Interoperability of Forensic Data — Interoperability of forensic data provides the basis for improved collaboration and sharing of information among forensic laboratories, practitioners, and different forensic methods and systems, including automated systems. This theme covers topics such as scientific interoperability studies and tests; documentary interchange standards, practices

and protocols; and conformance tests for documentary standards.

4. Automated Forensic Technologies — Automated technologies can greatly enhance forensic practices by providing greater accuracy, reliability, repeatability, functionality, and throughput. This theme covers topics such as measurement and evaluation approaches for automated technologies; technologies that perform forensic information extraction, representation, and analysis; software tools to measure forensic data quality; forensic database representation and search; digital forensic technologies that extract and analyze digital data; and usability of automated technologies.

Measurement Science, Statistical Foundations, Automated Technologies

A192 Mitigating Risk in the Forensic Drug Chemistry Laboratory

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After attending this presentation, attendees will learn of key measures to reduce the chance of analytical errors and the opportunity for intentional misconduct in a forensic drug chemistry laboratory.

This presentation will impact the forensic science community by showing specific responses to well-known lapses of laboratory integrity in the hopes of preventing these occurrences in other laboratories.

By studying the policies and practices of laboratories that have experienced “dry labbing” and/or data falsification incidents, it is possible to identify a series of risk factors that are likely to foster these inappropriate behaviors that may occur unnoticed. Six key factors that should be monitored in order to mitigate the risk of malfeasance are: the opportunity for forgery/falsification; failure to perform analyses; unverified exceptional productivity; lack of physical visibility; inappropriate communication with customers; and ineffective supervisory responses when potential issues arise.

Forgery/Falsification: As chemists inevitably become acquainted with co-workers’ signatures and handwriting, it may be tempting to forge initials on instrument maintenance logs or other data in order to “improve productivity.” However, the implementation of PIN-protected signatures through a Laboratory Information Management System (LIMS) system, or document viewer, eliminates this scenario. In order to guard against chemists claiming credentials they do not legitimately possess, training certificates, as well as copies of all degrees, should remain on file and be verified annually.

Failure to Perform Analyses: A primary method of ensuring that analysis is truthfully performed is through the implementation of analytical processes that provide reviewable data. This includes all instrumental data as well as photographs of microscopic botanical characteristics and color tests. Traceable balance software can be implemented that not only captures sample weight data, but prevents any weighing event from occurring if the required calibration check was not successfully performed. In order to prevent casual access to analytical standards, which may be used to spike a drug sample, access should be minimized. Additionally, non-traditional case workflows may be implemented to increase quality assurance levels as well as increasing the number of individuals involved in a single case.

Unverified Productivity: Laboratory and individual

productivity should be monitored on at least a monthly basis, and the activities of staff with an abnormally high or low productivity should be scrutinized. An analyst with exceedingly high output should be closely monitored. Basing promotions or incentives on productivity alone creates an environment which promotes “cutting corners” in analysis.

Lack of Visibility: While it may not always be possible to modify the physical layout of the laboratory, providing an analyst with an isolated environment with no accountability is ill-advised. Locating all analysts in the same location prevents any type of secrecy. Additionally, the installation and active monitoring of security cameras provides both a deterrent to misconduct as well as a means by which questionable incidents may be readily reviewed.

Inappropriate Communication: In order to best avoid bias, policies should be in place — and actively enforced — such that laboratory analysts have minimal contact with customers, and that any contact that does occur is properly documented. If routine contact with a law enforcement or judicial agency is required, it is recommended that non-analytical staff be utilized for these communications.

Effective Supervision: In any laboratory environment, competent, effective supervision of analysts is critical. Policies should be in place that encourage employees to be aware of, and report, any suspected misconduct. When such reports are received, it is imperative that management follow up with a prompt investigation, potentially requiring the temporary removal of an analyst from casework while the investigation proceeds, as a precautionary measure.

The measures listed above, and others, have been implemented over time at the Harris County Institute of Forensic Sciences with minimal impact on turnaround time (after an initial acclimation period), in an effort to protect the integrity of the submitted evidence, the data the laboratory produces, and the chemists of the laboratory.

Dry Labbing, Data Falsification, Risk Management

A193 Reducing Backlogs as a Systems Problem

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After attending this presentation, attendees will have knowledge of how a systems-thinking approach can aid in the reduction of case backlogs (unworked cases older than 30 days).

This presentation will impact the forensic science community by providing a novel, conceptual approach to backlog reduction in forensic laboratories and, more importantly, instituting systems to keep backlogs reduced and managed.

The current but waning paradigm of machine-age thinking, where mechanisms or problems are taken apart like a machine to understand how the immediate components work, fares poorly in the face of ever-increasing and more-connected information. Individual parts no longer equate to the larger whole, the system, the environment, and externalities are as important and dynamic as the thing under study. Systems thinking takes the perspective that systems come about as a result of the interactions and relationships among their elements. These interactions, and

their emergent behaviors and unintended consequences, are as much a part of the system as the individual components.¹ Systems cannot be divided into independent parts; therefore, every element of a system loses some properties if removed from the system and the system — as a whole — has essential properties that none of its elements do.²

Problems can be thought of as a system with a critical value proposition attached.³ A problem has an external context and internal structure. It should be addressed through a process of systemic inquiry, such as Checkland’s Soft Systems Method (SSM).⁴ Backlogs at the District of Columbia Department of Forensic Sciences’ Forensic Science Laboratory Division (FSL) were addressed using this approach. Backlogs are hysteretic in nature (history dependent) and can be thought of as the cumulative lag in a system; this is true regardless of their rate of development. Predictable amplifications of small lags are a disproportionate cause of later circumstances; in the long run, this “historical hangover” leads to inefficiency, that is, backlogs.

Using relevant metrics from the FORESIGHT process, the following data was collected for CY13 (as of July 1, 2013):⁵

	Cases	Reports	Backlogged Cases
DNA	323	71	234
Fingerprints	1404	706	2379
Firearms	876	40	793

While case submissions exceed work capacity in each unit, evaluation of inputs and outputs did not necessarily equate to the backlogged values; streamlining of casework processes suggested achieving acceptable but not desired output rates. Other environmental factors had to play a role. Through a systematic review of court files, backlogged cases, service requests, and open investigations, significant backlog reductions were achieved. Therefore, the backlog derived not from an internal process but from a lack of a systematic feedback loop (communication) from external entities (attorneys and police) into the FSL and vice versa.

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Laboratory, Backlog, Systems

A194 Physical Evidence as a Quality-Control Tool in Criminal Justice

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After attending this presentation, attendees will learn that criminalistics and its focus on understanding the physical evidence record in events that become the subject of criminal proceedings has the potential for making contributions in three general areas. It is most commonly employed to support criminal prosecutions. To

a lesser degree, it can provide information of value in a criminal investigation. The third and perhaps most important area, although it may not be widely appreciated, is that of serving as a quality-control tool in criminal proceedings. This often-unnoticed contribution of criminalistics needs to be more widely recognized and appreciated.

This presentation will impact the forensic science community by raising awareness concerning the value of criminalistics and the physical evidence record as a quality-control tool in criminal justice processes.

Much has been written about wrongful convictions that were laid at the feet of faulty or inadequate forensic science. From our perspective, this seems to “turn the issue on its head.” In all of this, it is felt another important vantage point has been overlooked. In these cases, it is probably more accurate to see this as a failure for criminalistics to expose and correct earlier case-specific failings in the justice system.

Traditionally, criminalistics and its goal of extracting information from the physical evidence record are seen as having an investigative and an adjudicative role. Temporally, the criminalistics contribution to the investigation should take place as early as possible in the process and the contribution to the development of the prosecution case comes relatively late in the process. In addition to having a critical role in assisting with prosecutions and lamentably, somewhat less generally, solving crime, the scientific evaluation and interpretation of physical evidence should also be regarded as a quality-assurance tool for the criminal justice system. It should serve as a safety net in helping to prevent unsupported or erroneous convictions. This third, less recognizable role, which is temporally interwoven with the other two, appears to be an unappreciated contribution of criminalistics to the criminal justice system. Its invisibility does not diminish its importance. It is, in fact, critically important and needs to receive explicit recognition and support. Early in the investigation, it may imperceptibly manifest itself by subtly truncating false leads. Later, as a case gains momentum, if the need for corrective action is indicated, it becomes more difficult to make the necessary course correction, but the information provided by the physical evidence interpretation needs to be heeded. Properly recognized, utilized, and interpreted, physical evidence can provide the ground truth in understanding an event. How often does the physical evidence contradict or refute a prosecution hypothesis in the early stages and terminate a case? This appears to be unknown. Research to acquire data on this would be valuable.

It is rare for a single action to result in an adverse outcome such as a wrongful conviction. The physical evidence record can be the “canary in the coal mine” or sentinel providing an alert that something earlier in the process has gone wrong. Because much of this contribution may take place “under the radar” very early in the process, before much has been invested, its value is especially important, but may escape attention at the time. If employed earlier in the investigation, it can nip false leads in the bud. In the later stages of a case, it can correct errors that have gained momentum, if it is appreciated and utilized to its fullest.

A limited number of case examples will be used to illustrate several points.

Physical Evidence, Quality of Justice, Justice Safety Net

A195 Should Forensic Sciences Only Be Confined to Court Use? The Emergence of Forensic Intelligence-Led Policing

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After attending this presentation, attendees will understand how forensic sciences can be used to generate intelligence in order to lead police investigations.

This presentation will impact the forensic science community by reminding everyone that the original purpose of forensic science is to help police investigate and by providing the knowledge necessary to do so.

Nowadays, forensic sciences work is most often court-oriented, meaning that all examinations are conducted with the mindset that they could end up in court on behalf of or against a defendant; however, forensic sciences can also be used to generate intelligence. Some clear practical examples will be provided of how to use forensic sciences to generate intelligence in place of or before going to court.

In the court-oriented mode, forensic sciences are used to identify a perpetrator or a victim or to determine a *modus operandi*. Examinations are almost always conducted with the mindset that the forensic scientist may have to testify about the results in court; therefore, the scientific method and conclusions must be beyond reproach. Protocols are verified, results are double-checked, and the conclusions must be error-free, or at least exhibit a known error-rate. Basically, when a forensic scientist works in court-oriented mode, one should be able to rely on the conclusion of the report without any reservations. These are the forensic sciences everyone knows about and performs.

In intelligence mode, forensic sciences do not have to provide the same level of accuracy or reliability. Because the investigator has no lead in the investigation, any information provided may help to find other clues, identify a lead, and eventually resolve the case. This occurs, for example, when the forensic scientist informs the investigator that the fibers may come from a certain car model, without being sure at all about this conclusion. It helps the investigator to target the search, without any guarantee that this is truly the car model sought. If the investigator follows a wrong lead, no harm is done besides the loss of time. As such, even if the intelligence provided is only 60% or 70% accurate, it is better than no intelligence.

This intelligence mode has undergone tremendous development in Switzerland. The main reason is that Switzerland exhibits a very high volume of burglaries, a serial crime occurring across many different states. Time is of the essence in detecting the series, because these burglaries are committed by foreign nationals who are often illegals merely transiting through the country. When sharing forensic data such as shoe prints or *modus operandi*, investigators can quickly link different burglaries and detect a series. This, in turn, allows for better tactical decisions on police operations. It is called forensic intelligence-led policing.

Efforts have been made in the last few years to redistribute crime scene unit resources in order to increase the production of forensic intelligence, particularly in response to high-volume crimes such as burglaries. Databases of forensic evidence have been developed and are used on a daily basis to provide guidance to the investigator and information to the decision maker.

While the focus of forensic sciences has been solely based on court-oriented practices for the last few decades, one must remember that the spirit of forensic sciences was not originally limited to this purpose. It was first developed not only to prove facts in court but also, maybe even more importantly, to further the criminal investigation by providing leads. It is crucial that modern forensic scientists always keep performing their duties with these two visions in mind. This can only increase the value of a crime laboratory's services to the investigator.

Forensic Sciences, Intelligence, Police Investigation

A196 Statistical Aspects of the Forensic

Source Identification Problem

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After attending this presentation, attendees will have a greater understanding of the current trends in statistical evidence interpretation, which will foster better communication between statisticians, evidence interpretation experts, and the broader forensic science community.

This presentation will impact the forensic science community by improving communication between these experts and should assist in the development of statistically sound, rigorous methods of interpretation that are appropriate to the diverse needs of the U.S. forensic science and legal communities.

In 1977, Dennis Lindley, with Ian Evett, introduced modern Bayesian methods for forensic evidence interpretation to the forensic science community. This and related approaches have dominated the academic research related to the interpretation and presentation of forensic evidence; however, in recent years there have been number debates, in both academic circles and forensic communities, related to the applicability of these methods in the U.S. judicial system.

Broadly speaking, these methods require the explicit statement of two mutually exclusive, but non-exhaustive, propositions about how the evidence in a given situation has arisen; one usually corresponding to a defense proposition and one corresponding to a prosecution proposition. Using this approach, once these propositions have been defined and the evidence has been collected, the forensic science expert is then required to present the evidence in a concise and transparent manner so that a decision maker can ultimately decide between the two proposed models of how the evidence has arisen.

Commonly, the evidence that a forensic scientist has available to evaluate between the two propositions is generally one of the following forms: (1) a trace of unknown origin; (2) a sample from the specific source specified by the prosecution hypothesis; or, (3) a collection of samples from the alternative source population specified by the defense proposition. In certain applications, the choice of the alternative source population will be mandated by available databases or, in extreme situations, there will be no such samples available.

This presentation, will review some of the common sets of propositions and statistical approaches that forensic scientists use to characterize the support that the evidence provides for deciding between the prosecution and defense propositions. Discussion will also include how the various sets of competing propositions can be addressed with the commonly available evidence. The general approach will be illustrated with examples that arise in trace element analysis of high purity copper and glass evidence.

Statistics, Evidence Interpretation, Trace Elements

A197 Transparent Latent Print Testimony: When the Basics Are No Longer Basic

Heidi Eldridge, MS, LVMPD Forensic Laboratory, 5605 W Badura Avenue, Ste 120 B, Las Vegas, NV 89118-4705*

After attending this presentation, attendees will be acquainted with the need to articulate basic concepts in fingerprint comparison science in a way that is modest, transparent, and helpful to a trier of fact. Many examiners have expressed frustration in the wake of the National Academy of Sciences (NAS) Report that they

are not supposed to say things like “zero error rate,” “exclusion of all others,” and “100% certainty,” but they don’t know why not and they don’t know what to say instead.¹ This presentation will provide that explanation and guidance, showing attendees a better way to express their findings in court.

This presentation will impact the forensic science community by providing examiners with a roadmap to more transparent testimony and helping them to understand the underlying concepts in the current paradigm shift away from claims of uniqueness and global individualization. This adjustment is not without its growing pains and may be downright intimidating to some. This presentation will break down these daunting concepts into simpler and more defensible pieces.

Once upon a time, latent print examiners knew exactly what to say in court. A latent print is a representation of the friction ridge skin that has been transferred to a surface by touch. Because friction ridge skin is unique and permanent, that representation can be matched back to its source skin with 100% certainty to the exclusion of any other donor in the world. If the process is followed correctly, there is no possibility of error, making latent print evidence the most reliable form of identification there is.

Life was simple back then. Unfortunately, the way fingerprint evidence has traditionally been presented in court is problematic. Uniqueness theory has not been (and never will be) empirically proven. Global individualization is an inferential leap that is conceptually unnecessary. Errors have happened, making claims of a zero error rate demonstrably false, particularly as it makes no sense to separate method error from practitioner error in a process where the practitioner *is* the instrument. Absolute certainty is an unscientific concept that should not be espoused by anyone claiming to be a scientist or to be using a scientific process.

All this begs the question: what *should* latent print examiners say? Suddenly, all the comforting phrases that were so carefully learned during training are taboo and nobody has explained why, or told you what you *can* say. What do you do when everything you thought was a basic tenet of your science is now off-limits?

This presentation will explore the concepts of uniqueness, specificity, discriminability, error rates, analyst variability, certainty, and the identification decision in the context of the relevant population. We’re going back to basics and reinventing ourselves in a new, modest, transparent, and scientifically defensible way. You’ll learn that you really don’t need grandiose claims about individuality and infallibility to be able to present probative, reliable evidence in court. The jury will still trust you, the judge won’t limit your testimony, and you won’t lose your job to a computer spitting out probabilities.

In this presentation, you will learn how to articulate your decisions and lay the foundations for your science clearly and defensibly (not defensively!), with current research to back you up.

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Latent Prints, Testimony, Transparency



Digital & Multimedia



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B1 Experimental Analysis of the Changes in Pre- and Post-Operative Cosmetic Surgery (Rhinoplasty) by Comparison of 3D Images

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The goal of this presentation is to provide a tool based on scientific and innovative 3D reconstruction useful for an objective comparison between features of the faces in the pre- and post-operative period; in particular, the study focused on changes in the characteristics of the nose after surgery.

This presentation will impact the forensic science community by showing the usefulness of the reconstruction and analysis of 3D models pre- and post-surgery, allowing attendees to view, in an objective manner, changes of the nasal morphology.

This study first obtained the consent of the participants consisting of eight patients aged between 23 and 40 years old, who had chosen to undergo cosmetic surgery of the nose. Three acquisitions were made by the camera: on the pre-operative day, seven days, and 90 days after surgery. The stereo photogrammetric system used consists of two 3D cameras with a resolution of ten megapixels, a dual optical zoom of 3X and maximum picture resolution of 3888 x 2592. On the 3D images of the faces, landmarks were set to study the morphology of the nose (nasal, pronasal, subnasal, upper lip, left and right wing). Then the distances, indices, angles, volume, and the nasal area were calculated. This study focused on the pre- and post-operative values at 90 days; post-operative values detected a short time after surgery were not used because of the influence of inflammation of the soft tissues. Changes can be represented *qualitatively* by analyzing the color maps and *quantitatively* by numerical extrapolation of differences in measures, volume, and surface of the noses compared. Statistical analysis has also allowed assessment of how the measurements of the nose varied after a rhinoplastic surgery. This study showed that the width of the nose, the length of the nose bridge, and the height and the length of the nasal philtrum tend to vary little after rhinoplasty and thus can be taken into account for the possible recognition of a subject accused of a crime who undergoes cosmetic surgery of the nose. This comparison can be an effective tool in medical-legal cases: the professional responsibility of the plastic surgeon allows one to assess qualitatively and quantitatively the differences of the nose, frequently the subject of the claim. The comparison of 3D models pre- and post-surgery allows an objective view of the changes in

nasal morphology. These data represent an important proof that the consultants can present in court on the basis that this comparison is made between 3D models perfectly aligned with each other and overlapping. The study also has an interesting application in the context of personal identification; in fact, the analysis of changes in the morphological characteristics of the face, determined by a rhinoplasty surgery, allows one to define the difference between a subject filmed before plastic surgery of the nose and a suspect that has deliberately decided to fake his or her appearance. The results of this study encourage an expansion of the search.

3D Images, Rhinoplasty, Surgery

B2 Forgery Detection From Printed Images: A Tool in Crime Scene Analysis

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After attending this presentation, attendees will understand the risk of image manipulation and the tools for forgery detection.

This presentation will impact the forensic science community by focusing on the importance of forensic tools in image authenticity.

The preliminary analysis of the genuineness of a photo is the first step of any forensic examination that involves images, in cases where there is not a certainty of its intrinsic authenticity.

Digital cameras have largely replaced film-based devices. Until recently, in some countries, only images made from film negatives were considered fully reliable in court. There was widespread prejudicial thought regarding a digital image which, according to some people, could not ever be considered legal proof, because of its "inconsistent digital nature."

Great efforts have been made by the forensic science community in this field and now different approaches have been unveiled to discover and declare possible malicious frauds in order to establish whether an image is authentic or not or, at least, to assess a certain degree of probability of its "purity."

In this day and age, it's an easy practice to manipulate digital images by using powerful photo-editing tools. In order to alter the original meaning of the image, copy-move forgery is the one of the most common ways of manipulating the contents. With this technique, a portion of the image is copied and pasted once or times elsewhere into the same image to hide something or change the real meaning of it.

Whenever a digital image (or a printed image) will be presented as evidence in a court, criteria should be followed to analyze the document with a forensic approach to determine if it contains traces of manipulation.

Image forensics literature offers several examples of detectors for such manipulation. Among them, the most recent and effective are those based on Zernike moments and those

based on Scale Invariant Feature Transform (SIFT). In particular, the capability of SIFT to discover correspondences among similar visual contents allows the forensic analysis to detect even very accurate and realistic copy-move forgeries.

In some situations, however, instead of a digital document, only its analog version may be available. It is interesting to ask whether it is possible to identify tampering from a printed picture rather than its digital counterpart.

Scanned documents or recaptured printed documents by a digital camera are widely used in a number of different scenarios, from medical imaging and law enforcement to banking and daily consumer use.

In this presentation, the problem of identifying copy-move forgery from a printed picture is investigated. The copy-move manipulation is detected by proving the presence of copy-move patches in the scanned image by using the Copy-Move Forgery Detection (CMFD) method; previous methodology has been adapted in a version tailored for printed image case (e.g., choice of the minimum number of matched keypoints, size of the input image, etc.).

A real case of murder is presented where an image of a crime scene, submitted as printed documentary evidence, had been modified by the defense advisors to reject the theory of accusation given by the prosecutor.

The goal of this presentation is to experimentally investigate the requirement set under which reliable copy-move forgery detection is possible on printed images in such a way that the forgery test is the very first step of an appropriate operational checklist manual.

Forgery Detection, Copy-Move Tampering, Crime Scene Investigation

B3 A Comparison Between the Windows® 8 & Windows® 7 Registries

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WITHDRAWN

B4 Comparison of Super Resolution Image Enhancement Algorithms for Forensic Image Analysis

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After attending this presentation, attendees will gain an understanding of some principles of multi-frame Super-Resolution (SR) image enhancement techniques; various SR algorithms have been proposed, quality of the resulting images are different depending on the algorithm, and the optimum algorithm depends on the type of processing images.

This presentation will impact the forensic science community by providing the experimental results showing the best SR algorithm varies depending on the nature of source images. Even if image quality is not improved with a specific SR algorithm, there is a possibility of obtaining a better resulting image by using

a different algorithm. This knowledge will be useful for forensic image analysts who think that the multi-frame SR techniques are not functioning well for real-case images.

Videos or images that were recorded by surveillance or any other cameras can be objective evidence leading to criminal investigation. However, in many cases, the recorded images cannot be effectively utilized due to insufficient spatial resolution of the image. For such image degradation factor, multi-frame SR processing is highly effective for image quality improvement: it is possible to improve the spatial resolution of the image by integrating information from many images. Various algorithms have been proposed for multi-frame SR, and their effectiveness has been shown in the literature.^{1,2} However, from the experiences of some professionals, it did not work well to most actual case videos even though clear results are shown in academic papers.

This study will present experimental results of the performance of various multi-frame SR algorithms using various image sequences including images taken by surveillance cameras. For the experiments, more than five registration algorithms (Keren, LK, Farneback, Brox, Dense LK, Simple Flow, etc.) and five reconstruction algorithms (Interpolation, POCS, NC, BTV, MAP) were implemented; usually multi-frame SR processing is performed in two steps (registration and reconstruction). These SR algorithms were written in C++ language and they were implemented using the computer vision library OpenCV 2.4. For the combination of these algorithms and input image sequences, the resulting image quality was assessed. The input image sequences included both computer-generated and camera-recorded image sequences.

The results show that some surveillance camera images can be improved with the SR image processing under a specific condition so as to identify unknown characters. However, in comparison with the images that were generated in a computer, image quality improvement was lower for the camera recorded images. Furthermore, it was found by the experiment that the optimum algorithm was different from the source images. The reason for this was assumed to be due to the difference of the image observation model that each algorithm hypothesized. Therefore, from the aspect of the actual image analyses on crime investigations, it is considered important to implement various SR algorithms and to select the optimum one according to the source video; because a wide variety of evidentiary materials are treated in the forensic activities.

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Image Enhancement, Super Resolution, Algorithm

B5 Random Access Memory Persistence: When Does It Go Away?

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After attending this presentation, attendees will understand some basic principles and behaviors related to the persistence of computer random access memory.

This presentation will impact the forensic science community by providing a basic understanding of what forensics artifacts can be found in Random Access Memory (RAM) data captures and what affects its persistence when a computer is shut down, power is removed, or other events occur that can affect RAM.

RAM is known to potentially contain many forensic artifacts related to investigations such as incident response, child

exploitation, and almost all other computer forensic cases. These artifacts can include evidence such as images or partial images, malware code or partial malware code, passwords or password hashes, and words used in a variety of computer applications.

There have been a number of articles written over the years about capturing and analyzing RAM. Indeed, there are several groups providing week-long “introductory” classes in RAM capturing and analyzing. There are also articles published now on when RAM may be still available for capture when, for years, assumptions were made that RAM would be cleared, such as when computers are shut down.

This presentation will examine scenarios when RAM appears to persist after shutdown, re-boot, and removal of power. Testing is done where RAM is captured when it is known to be clear after using the computer in a variety of shutdown scenarios including, but not limited to: normal shutdown; pulling the plug; normal shutdown followed by pulling the plug; those scenarios and removing the RAM modules from the computer; etc. These tests are also performed on a laptop computer which adds the element of battery power to the above scenarios.

Possible complicating issues are examined including data that is cached on storage media that can still be analyzed for data from RAM, but also may populate captures from some or all data capture utilities. This cached data is compared to RAM captures. Further tests are done with the cached files eliminated to verify that what is seen in the RAM capture is strictly from volatile data and not cross-populated with non-volatile stored data from memory cache files.

The need for additional research and ideas about future research in this area of forensics will be presented.

RAM, Memory, Random Access Memory

B6 Querying Different Configurations Simultaneously to Conduct Database Forensic Examinations

Martin S. Olivier, PhD, University of Pretoria, Computer Science, Pretoria 0002, SOUTH AFRICA*

The goal of this presentation is to explore the possibilities of simultaneously querying multiple configurations of a database for forensic purposes. A configuration here is defined as a particular combination of selected American National Standards Institute, Standards Planning and Requirements Committee (ANSI SPARC) intension-extension layers. The presentation will demonstrate the power of such queries and recommend implementation strategies.

This presentation will impact the forensic science community by extending database forensics — an area where very little research exists.

It is common practice to, where possible, examine digital evidence in a “clean” environment. This is typically achieved by imaging the media and examining the image in a laboratory or by booting the untrusted device with a known (clean) operating system — often from a read-only medium. In previous work, it was demonstrated that the notion of a clean environment in the database forensics context is ambiguous given that the data in the database is interpreted through layers of metadata from which the data derives meaning.¹ In previous work it was also demonstrated that different configurations of clean and found layers may be assembled to be able to best answer the forensic questions of interest.² However, it is obvious that there will not always be a clear “best” configuration. In fact, often hypotheses will be best tested by asking questions about differences in query results provided by different configurations. The thesis in earlier work was that normal

query languages (such as SQL) provide a powerful mechanism to obtain evidence (or leads) from a database. The current work claims that such query languages are even more useful in the case where different configurations are examined simultaneously and proposes that this technique should be explored in detail.

In order to formulate queries across configurations such, configurations need to be amalgamated. Two primary options exist: either the various configurations need to be integrated (in a way similar to normal schema integration), or the configurations have to be configured as a distributed database. In the latter case, a number of alternatives exist (which may be systematically explored using Özsu and Valduriez’s taxonomy of such databases).³

It is demonstrated that the ideal approach for such comparisons depends on the configurations that are to be processed. Where the data models or data dictionaries of the configurations differ, a distributed architecture is indicated. It will be shown that a federated architecture is the best general solution, based on the fact that nodes in a federated database retain autonomy and user permissions will form an inherent part of many database forensic examinations. However, other distributed database architectures may be better suited for specific categories of analyses.

In contrast, where the data model and data dictionary are consistent among the configurations to be examined, schema integration is indicated. (In this case, configurations will differ on the schema and/or data layers — possibly in a temporal dimension.) Schema integration is facilitated by the fact that the configuration schemas will be identical or very similar in these cases, and a simple tagging approach may be used to distinguish artifacts from the different configurations. Such integration allows a more natural use of the query language (that will facilitate communication of results to non-technical parties in a case). In contrast, the distributed database approaches indicated in the other cases call for a more complex query language. In the case of SQL, in particular, extensions to the language may be required to adequately express forensic queries where configurations are combined as a distributed database.

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Database Forensics, Forensic Queries, Forensic Configurations

B7 Compressed RAM in Linux® and Mac® OS X®: Impacts on Live Forensics

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After attending this presentation, attendees will understand the key characteristics of modern virtual memory systems that impact memory analysis in live forensics investigations, with a focus on emerging “compressed swap” facilities. Despite the decreased costs of RAM upgrades and growing memory capacities of modern computer systems, there is substantial interest among

operating systems developers in improving utilization of physical memory. There are various reasons for this, including the popularity of extremely portable and relatively resource-poor ultrabooks and pervasive use of virtualization. Compressed RAM facilities will “break” current generation memory analysis tools, and while modification of existing tools will not necessarily be trivial, there are potentially great rewards in doing so. This presentation will survey the compressed RAM facilities in both Linux® and Mac® OS X® and discuss the impact on both memory analysis tools and capabilities.

This presentation will impact the forensic science community by providing an accessible introduction to virtual memory system internals, the structure of current generation memory acquisition and analysis tools, emerging mechanisms for compressing RAM to increase performance and decrease swapping in Mac® OS X® and Linux®, and the impact these mechanisms will have on memory analysis. This work is important because virtually all memory acquisition tools will require adaptation to return complete and correct results in the presence of compressed RAM.

Historically, efforts to compress RAM to make more memory available for applications has had limited success. Applications like RAM Doubler for Mac® OS X® were popular decades ago, yet improved system performance only under a restrictive set of circumstances. The advent of modern multicore processors is providing new life for RAM compression mechanisms, which will advent in both Mac® OS X® Mavericks (Mac® OS 10.9) and in newer versions of the Linux® Mac® Mac® kernel. The goal of these mechanisms is to better utilize physical memory resources, reduce swapping to hard drives/SSDs, and improve system performance. Despite memory capacities increasing and the cost of memory upgrades decreasing, these facilities make sense in a number of circumstances, because even with modern SSD designs in desktop systems, bandwidth is capped at just over 1GB/sec, while physical memory bandwidth may reach 60GB/sec or more, making swapping extremely expensive.

The deployment of these compressed RAM mechanisms will break virtually all current-generation memory acquisition and memory analysis tools. Further complicating this issue is that the facilities in Mac® OS X® and Linux® are different and that Linux® will actually offer several alternatives for deploying compressed RAM. Memory analysis tools rely on accurate and complete physical memory acquisition and with the introduction of compressed RAM, the relatively platform-independent methods currently used for acquiring RAM on Intel-based systems will now require OS-specific (and compressed RAM mechanism-specific) techniques for acquisition, substantially increasing complexity. The benefits of compressed RAM for live forensics analysis, however, may far outweigh the effort in fixing tools, since memory pages that were previously swapped out (and therefore not analyzable by most memory analysis frameworks) may actually be present in the compressed areas of a memory capture.

Digital Forensics, Memory Analysis, Compressed RAM

B8 Kernel Pool Monitoring to Support Malware Forensics in a Cloud Computing Environment

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After attending this presentation, attendees will understand the internals of dynamic memory allocations (a.k.a. kernel pools) in Microsoft® (MS®) Windows® and how virtual machine introspection can be used to monitor the pools in a cloud-computing environment for detecting malware infection. Malware typically modifies function pointers to redirect the control flow of the system to execute malicious code. Tracing and identifying such stealthy hooks has significant importance in malware forensics. This presentation particularly focuses on monitoring and logging any suspicious modification of function pointers in the kernel pool to facilitate post-incident malware forensics.

This presentation will impact the forensic science community by discussing a method of monitoring and logging suspicious modification of function pointers in kernel pools. The method is unique in that it works on MS Windows® — a high priority target for malware developers, and only relies on the artifacts residing in the physical memory of the target system to work. It is also the first system of its kind that allows both 32- and 64-bit versions of the Windows® kernel to be monitored for function pointer integrity.

Microsoft® has introduced kernel patch protection or PatchGuard to protect kernel code and important data structures such as the Interrupt Descriptor Table (IDT) that are typically targeted by traditional malware. This has made it difficult for malware developers to exploit the code and data structures protected by the PatchGuard. Thus, they look for low profile targets for exploitation in unexplored regions. The kernel pool is one such region where, in particular, function pointers are targeted to execute malicious code. There are numerous function pointers in the kernel pool, which provide an attractive opportunity for an attacker to install stealthy hooks. State-of-the-art solutions perform static analysis of kernel source code to obtain precise definitions of kernel data structures, which also identify the locations of function pointer fields in the data structures. Moreover, they generate traversal graphs by linking pointers from one data structure to another, which is then used to target the data structures in the physical memory of a target system for integrity checking. Since the solutions require source code for the operating systems kernel, they are generally not applicable to MS® Windows®. The seminal work of Yin et al. on MS® Windows® uses taint analysis to learn about the definition of and contextual information for kernel data structures.¹ However, it is dependent on taint analysis for accuracy. The work is also dependent on the relocation table to track function pointers in binary code. However, the relocation table does not contain entries to locate function pointers in 64-bit Windows®, which limits the applicability of their work to 32-bit versions of Windows® only.

This study's approach works in a virtualized environment and identifies function pointers that are maliciously modified in a kernel pool. It runs in a privileged virtual machine and uses virtual machine introspection to access the physical memory of a target Virtual Machine (VM) running MS® Windows®. Since it runs outside the target VM, it is less prone to subversion if a target VM is compromised. It obtains the list of function pointers directly from reliable sources in the physical memory of the target machine, without relying on source code, disassembling the kernel binary, or traversing the relocation table. The pointer list is then used to find the instances of function pointers in kernel pool data

for integrity checking. It does not require hooking to obtain the kernel pool data; instead, it uses kernel data structures maintained by Windows® to track memory allocations to locate appropriate dynamic allocations in kernel pools. This study has implemented the technique and details will be discussed in the presentation. Our experimental results show that a small region in the kernel pool has a high concentration of function pointers, which is also non-pageable, providing a rich reliable attack surface for exploitation. The implemented tool is able to perform real-time monitoring of the region.

Reference:

1. H. Yin, P. Poosankam, S. Hanna, and D. Song, "HookScout: Proactive Binary-Centric Hook Detection", in Proceedings of the 7th International Conference on Detection of Intrusions and Malware, and Vulnerability Assessment (DIMVA'10), Bonn, Germany, 2010, pp. 1-20.

Cloud Computing, Integrity Checking, Kernel Pool

B9 Intelligently Combining Carving Results to Reduce Examiner Workload and Improve Output

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After attending this presentation, attendees will understand why combining carving results can be useful and understand some of the many issues combining these results can create. Discussion will include some of the more common carving tools with respect to accuracy (false positive/false negative results). Current activities across the community to identify intelligent ways to combine and display the results will also be shown. Finally, the potential issues that solid state devices may have on carving will be discussed.

This presentation will impact the forensic science community by showing the research that has been done in this area over the preceding months and the potential approaches to make this work.

The digital and multimedia forensics community is always looking for ways to improve processes. Improvements in carver results such as: reduction of false positives, false negatives, and greater accuracy can reduce examiner work load and improve accuracy of results. Combining the results of multiple tools, selecting the best results, and discarding the junk should improve overall results. The problem with this approach is knowing what is good and what is bad.

The research presented assumes that different carving tools perform better with some file types than with others. Based on this assumption, with appropriate testing we should be able to improve overall performance by intelligently combining the result of multiple carving tools. Performance attributes of some specific commercial and open source tools will be discussed.

Combining the output of multiple tools is not simply intermixing the results and displaying them. This simple approach will likely increase the number of unique files found and will also likely drastically increase workload for examiners because of the massive amount of duplication that will occur. In addition to the obvious issues with duplication, there are significant less-obvious obstacles to reducing work load for examiners. As an example, many carving tools are said to be "better" than others because they have low false positive rates on some file types. If one combines the better carver with the results from poorer carvers (with higher false positive rates), the bad tool will simply add back in the false positives that the better tool rejected. So, intelligently combining these results may mean ignoring part of the output of each tool and only keeping the best of the results.

In addition to combining the outputs from the different carvers, this study looked at workflow and ways to improve carving efficiency by making a number of passes, removing items found and only carving the remainder.

Finally, this presentation will show the research that has been done in this area over the preceding months and potential approaches to make this work.

Carving, Tool Integration, Improved Output

B10 Mobile Device Tool Testing

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After attending this presentation, attendees will be aware of the importance of tool testing and will have gained an understanding of the mobile device tool testing process conducted within the Computer Forensics Tool Testing (CFTT) project.

This presentation will impact the forensic science community by increasing awareness of the role of tool testing in informing the forensic community of tool capabilities and limitations. Test reports provide a foundation for toolmakers to improve tools, help users to make informed choices, and provide interested parties with an overview of any anomalies found. This presentation will provide an overview of the motivation behind testing mobile device forensic tools and the challenges faced by toolmakers and forensic examiners.

The CFTT project has spent several years researching and testing forensic tools capable of acquiring data from the internal memory of mobile devices and Subscriber Identity Modules (SIMs). This presentation discusses all aspects of the testing process that are critical for producing a test report.

The development of mobile device forensic tools and acquisition techniques continues to grow within the field of digital forensics. Mobile subscribers far outnumber personal computer owners and studies have shown an increase of mobile device personal data storage compared to personal computers. Higher-end mobile devices present users with advanced features and capabilities similar to those of a personal computer. Mobile devices provide users with the ability to maintain contact information, upcoming appointments, day-to-day activities, important news events, and provide the ability to correspond with friends and family via telephony, text message, email, chat, and social networking sites. Over time, mobile devices can accumulate a sizeable amount of information about their owners. Data acquired from these devices may be useful in criminal cases or civil disputes.

As mobile device usage and sophistication continue to grow so does the need for tool validation. For acquired information to be admissible in a court of law, verification of a tool's behavior and strict forensic acquisition methods are paramount. Potentially, one piece of data acquired from a mobile device may play a critical role in shedding light on an incident or possible criminal activity. The need for rigorous testing conducted on a combination of forensic tools and specific families of mobile devices is critical for providing law enforcement and forensic examiners informative test results yielding known expectations of a tool's behavior, capabilities, and limitations. Over the past three years, the CFTT project at the National Institute of Standards and Technology has tested numerous mobile device forensic tools capable of acquiring data from mobile devices operating over Global System for Mobile (GSM) communications and Code Division Multiple Access (CDMA) networks.

This presentation covers information on the motivation behind testing mobile device forensic tools, specification and test plan development, creation of a known data set, mobile device data population, and tool testing.

This presentation gives an overview of the CFTT process as applied to mobile devices.

Mobile Device Forensics, Mobile Device Tool Testing, Computer Forensics

B11 Bluetooth® and Spyware Hacking of Android™ Smart Phones: A Forensic Assessment Using a Forensic Toolkit (FTK)

Marci L. McCloskey, MS, 1204 N Crawford Avenue, Norman, OK 73069; and Mark R. McCoy, EdD, Forensic Science Institute, 100 N University, Edmond, OK 73034*

WITHDRAWN

B12 Forensic Analysis of Data Transience Applications in iOS® and Android™

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After attending this presentation, attendees will understand the capabilities of data transience third-party applications and gain insight on potential factors that impact the recoverability of communication artifacts within the iOS® and Android™ device.

This presentation will impact the forensic science community by focusing on the recoverability of data from two popular third-party applications that might be used by criminals to circumvent the law. With mobile phones becoming a common piece of evidence to be investigated, recovered artifacts can potentially connect two individuals who otherwise would not be connected.

The availability of mobile applications has greatly enhanced the capabilities of mobile phone users. Among these applications are data transient apps such as Snapchat (Snapchat, Inc.) and Burner (Ad Hoc Labs, Inc.), which has become prevalent amongst mobile phone consumers. In Snapchat, users are able to share timed content that 'self-destructs' upon reaching the set duration, making it no longer accessible according to the privacy policies. The Burner application allows you to double a personal mobile phone as a burner phone, maintaining the privacy of the user. Upon expiration of those phone numbers, all history and logs associated with them are removed from the device.

Prefacing these applications with the ability of content termination, users may use these applications for the purposes of drug deals, distribution of child pornography, and other criminal activity, expecting any exchanged content to delete upon expiration. In these cases, the recoverability of artifacts becomes essential in investigations which includes, but is not limited to observing the transferred content, timestamps, and associations amongst individuals.

Using a LG® Nexus 4 E960 and a fourth generation Apple® iPod touch, a constant exchange of data was carried out. Physical extractions and file system extractions were conducted using the Celebrite® UFED Touch and the Celebrite® Physical Analyzer in order to study different factors impacting the recoverability in

Snapchat and Burner. Snapchat factors focused on message status, time elapsed, and the "clear feed" option while Burner factors focused on time elapsed and expiration method. The differences between the Android device and iOS device proved to be significant.

In a case requiring investigation of Snapchat data, time is of the essence when it comes to the Android due to the server ability to remove received snaps from the given accounts after a certain time has elapsed. The iOS device showed no recoverability of any snaps. Both devices showed logs, which no longer existed upon using the 'clear feed' option. The recovery of Burner application data, however, seems to be dependent upon whether the burner number was expired or manually removed.

This research was supported by Award No. 2010-IJ-CX-K025 awarded by the National Institute of Justice, Office of Justice Programs, U.S. Department of Justice. The opinions, findings, and conclusions or recommendations expressed in this publication/program/exhibition are those of the author(s) and do not necessarily reflect the views of the Department of Justice.

Snapchat, Burner, Mobile

B13 A Strategy for Testing Graphic File Carving Tools

James R. Lyle, PhD, NIST, 100 Bureau Drive, Mail Stop 8970, Gaithersburg, MD 20899; and Richard Ayers, MS, 100 Bureau Drive, MS 8970, Gaithersburg, MD 20899-8970*

After attending the presentation, attendees will be made aware of some of the issues revealed by creating and using data sets for testing computer forensic tools for carving deleted files from unallocated file space.

The presentation will impact the forensic science community by increasing awareness of tool test strategies and their ability to reveal anomalies in tool behavior. This presentation will aid the forensic practitioner in recognizing the limitations of file carving tools.

The Computer Forensic Tool Testing (CFTT) project at NIST develops methodologies for testing computer forensic tools. This presentation reports on creating data sets for testing file carving tools and their behaviors.

File carving is widely used in digital investigations to extract deleted files from unallocated storage. Usually file carving is applied to file types with a recognizable structure so that unallocated space can be scanned for file components that are then reassembled into complete files. If the file has easily identified beginning and ending content and is contiguously allocated then carving is simple. However, the reality of file fragmentation complicates the task considerably.

Categories of files that are common targets of file carving include:

- Graphics: JPG, GIF, PNG, BMP, TIF & PCX
- Videos: MP4, AVI, MOV, MPG, FLV & WMV
- Audio: MP3, WAV, AU & WMA
- Document: DOC, DOCX, XLS, XLSX, PDF, PPT & PPTX
- WEB: HTML, SQLite & chat
- Archive: ZIP, RAR, 7Z, GZ & TAR
- Misc: exec, logs, etc.

A common tool testing strategy is needed to help investigators characterize and understand tool behaviors, to compare tools, and to create test data with known content for investigator practice and training.

Data sets are available for download with each being available as a single file as would be created using the UNIX `dd` command to image a storage device.

1. A set of complete and contiguous graphics files with no intervening data between files – the last sector of each file is padded with zeros until the end of the sector. This test set reveals problems finding the beginning and the end of graphics files.
2. A set of complete and contiguous graphics files – the gap between files is filled with varied data in common cluster sizes, i.e., 1, 2, 4, 8, 16, 32, or 64 sectors. The gap content is varied over zeros, constant value, random data and various text formats. This test set reveals problems triggered by the presence of non-graphics files.
3. Files are placed such that each file does not begin on a sector boundary, but is offset from the sector beginning – this test differentiates algorithms that require the file to start on a sector boundary from those that can find an embedded graphics file.
4. Simple Fragmentation – a set of sequential fragmented graphics files. This test identifies how a tool deals with fragmentation.
5. Complex Fragmentation – the test set has some fragments out of order and some fragments intertwined. This test identifies algorithms that deal with complex fragmentation.
6. Incomplete Fragmented Files – some fragments are missing. This test set reveals complications when encountering incomplete files.

In this presentation, existing file carving test sets are examined to identify the underlying assumptions that have guided the creation of the test sets. From this, a new set of test images have been developed that can help investigators characterize and understand tool behaviors, compare tools, satisfy laboratory accreditation requirements and create test data with known content for investigator practice and training.

Certain trade names and company products are mentioned in the text or identified. In no case does such identification imply recommendation or endorsement by the author or the author's employer, nor does it imply that the products are necessarily the best available for the purpose.

The test images and image layout documentation are available at the CFRReDS project web site: <http://www.cfreds.nist.gov/FileCarving/>.

Test reports on specific tools are available from the Department of Homeland Security Cyber FETCH web site: <https://www.cyberfetch.org/>.

Digital, Software, Testing

B14 Tracking the Effects of Software on Systems: A Forensic Metadata Collection

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The goal of this presentation is to familiarize attendees with a data set available from the National Institute of Standards and Technology (NIST) and aimed at digital forensics investigators which tracks the lifecycle of targeted software and provides data on the files, Windows® Registry components, memory states, and network communications associated with the targeted software at specific points in its lifecycle, including after uninstallation.

NIST's National Software Reference Library (NSRL) is building a forensic data set which tracks the various effects of software on computer systems under controlled conditions,

providing forensics investigators with a baseline record of artifacts produced at specific points in the software's deployment.¹

Virtual machine (VM) environments built on common Windows® operating systems are used as installation environments for targeted software. At predetermined break points during the life cycle of the software, the VMs are paused and saved. Simultaneously, any network traffic originating from the VM is captured and is also saved at each break point in the life cycle. The software life cycle is defined as its installation, registration, online update, use and uninstallation. The sum of all data collected over the life cycle of the software is referred to as the software's "diskprint," while the data collected at each stage is referred to as a "slice." Once the software diskprint has been saved and recorded, the slices are processed and the associated files, Registry entries, RAM contents and network traffic are extracted for publication. Note that the baseline operating system installations were also processed in this manner: the data on clean operating system installations is also available.

The results are published in two formats: File metadata are folded wholesale into the NSRL RDS to supplement the file metadata garnered in the usual manner.² The file metadata, Registry information, RAM cells and network traffic are also published in an XML format that describes the diskprint in terms of its constituent slices.

NIST is working closely with the DFXML and the Cybox communities on the definition of a format for the publication of diskprints for easy ingestion into forensic tools.^{3,4}

The intention behind this data set is to provide investigators with definitive evidence of the artifacts associated with software packages, thereby reducing the community's reliance on untested knowledge, inference and hearsay and introducing a new level of rigor into the discipline.

References:

1. The National Software Reference Library is online at <http://www.nsrll.nist.gov>
2. See, for example, "NIST's National Software Reference Library": <http://www.nsrll.nist.gov/Documents/NSRLL-CFS-April-2009.pdf>
3. Digital Forensics XML in the Forensics wiki: http://www.forensicswiki.org/wiki/Category:Digital_Forensics_XML
4. Cyber Observable eXpression: A Structured Language for Cyber Observables: <http://cybox.mitre.org>

Diskprint, File System, Windows® Registry

B15 The National Software Reference Library: Applications in Digital Forensics

Douglas R. White, MS, 4225 Angell Road, Taneytown, MD 21787-2601*

After attending this presentation, attendees will be familiar with the data generated by the National Software Reference Library (NSRL), which is freely available to digital forensics investigators and researchers.

This presentation will impact the forensic science community by calling attention to the various data sets produced by the National Institute of Standards and Technology (NIST) NSRL.

The NIST National Software Reference Library has collected computer software since 2000. Acquisitions have spanned Microsoft®, Apple®, Linux®, and other operating systems. Applications range from common business software suites to foreign, malicious, specialized executables. The NSRL retains copies of all applications in the collection.

Each application is described with metadata, detailing the manufacturer, publisher, system prerequisites, etc. Every file in each application is described with metadata detailing the file name, path, dates, byte signatures, cryptographic hash (file fingerprint), etc. All of this metadata is available to the public. A subset of the metadata that is targeted for investigators is published quarterly and made available to the public.

All media in the collection are copied to network-based storage. All distinct files are stored in a corpus on network-based storage. "Archive" type files (Cabinet, Zip, tar files, etc.) are recursively extracted and content files are added to the corpus. As needs arise for heretofore uncollected metadata, NSRL can update and repeat processes to harvest additional information from the media and files. Access to media images and the file corpus may be obtained.

Software applications are installed in virtual machines to collect metadata on files, memory, and operating system structures (e.g., Windows® registry). Several points in the installation are chosen while the system is in a stable state, to enable identification of actions taken during the lifecycle of the applications.

Software acquisition includes mobile device applications, "clickwrapped" download-only applications, network-based multiplayer applications, and game console applications. The NSRL collaborates with other libraries and collections. Metadata taxonomies and standard software identifiers are used to enable information sharing between diverse data sets. A "Digital Forensics XML" (DFXML) schema has been drafted to facilitate discussion on interoperable data.

NSRL investigates methods of identification other than the use of cryptographic hashes. Block-level hashing introduces statistical probabilities into the process, and approximate matching involves algorithms that allow measurement of match characteristics.

File Identification, Digital Metadata, Registry

B16 Indexing the Windows® Registry for Software Detection

Alex J. Nelson, MS, 100 Bureau Drive, MS 8970, Gaithersburg, MD 20899-8970; Mary T. Laamanen, MS, NIST, 100 Bureau Drive, Gaithersburg, MD 20899; John Tebbutt, 100 Bureau Drive, STOP 8970, Gaithersburg, MD 20899-8970; and Darrell D. Long, PhD, 1156 High Street, Mailstop SOE 3, Santa Cruz, CA 95064*

After attending this presentation, attendees will learn about a technique to profile software on a Windows® computer to aid digital forensic triage. They will learn practical uses, investigative value, and structural characteristics of the Windows® Registry. They will learn a generic, information-retrieval-based measurement which can be used to attribute arbitrary components of the Registry to known software packages.

This presentation will impact the forensic science community by showing how being able to identify software usage on a computer is a boon to forensic triage. By detecting the software used on a computer, an investigator can receive key, rapidly actionable information, such as signs of malware or anti-forensic utilities. Searching for software presence, usage, and removal using the Windows® Registry allows one to translate what were purely machine-level artifacts back into human actions, illustrating the story of computers of otherwise unknown provenance.

The Windows® Registry is a central store for Windows® systems, recording configuration and system state. Forensic investigators find it a key resource in identifying system uses and reading specific records to support hypotheses about what a

computer was used to do.

Using a document search model, the Windows® Registry's extensive namespace allows one to identify software histories associated with a computer—including applications installed, run, and/or removed—with a measurable certainty.

The Registry is a data structure analogous to a specialized file system. It contains a hierarchical namespace used to store configuration values varying in size from as small as a byte to values in the kilobyte range. This namespace acts much like a file system, where its analogies to "files" and "directories" can be generally referred to as "cells." With baseline cell tallies of one hundred thousand in Windows® XP to almost a half-million in Windows® 8, there are many locations in which telltale signs of software activity can be left behind.

One strategy to record software effects is to install, use, and uninstall applications on virtual Windows systems by snapshotting virtual machines—a process referred to as "diskprinting" in the spirit of fingerprinting—and enumerating the differences. Differences in the Registry show the particular cells affected in each step taken, and these cells can be grouped into change sets. Then, a well-known information retrieval technique, document search, identifies applications likely to have affected arbitrary Windows® systems, along with the actions those applications took. Comparing the cells affected in each change set shows the distinctness of each as an in-Registry fingerprint. Distinct fingerprints enables one to identify, from an arbitrary Registry namespace, the signs of application presence, use, and removal. For instance, a standalone application, run not by installing but instead from a thumb drive, could show signs of the browser having run while signs of installation would be absent.

Registry cell paths are distinct to the combination of the operating system, application, and basic user action. A single application's installation, use, and uninstallation was observed to affect over fifty thousand cells in the Registry. Without resorting to analysis of memory or deleted content, the document search approach was able to identify software used in a research scenario that recorded real computers' state. Applications that had been diskprinted were recognized in the scenario, even though the version diskprinted was not the precise version used in the scenario.

The Windows® Registry is a file system, configuration store, and log, often a critical source in forensic investigations. This presentation shows that by looking not at the data stored in the Registry, but instead at the structure of this data, the software history of a computer can be learned. Some software effects in the Registry can be detected based on observing behaviors of an older version of the software. This implies that significant Registry patterns are preserved as products evolve, much like a genetic lineage.

Being able to identify software usage on a computer is a boon to forensic triage. By detecting the software used on a computer, an investigator can receive key, rapidly actionable information, such as signs of malware or anti-forensic utilities. Searching for software presence, usage and removal using the Windows Registry allows one to translate what were purely machine-level artifacts back into human actions, illustrating the story of computers of otherwise unknown provenance.

Windows® Registry, Information Retrieval, Differential Analysis

B17 Relatively Permanent Pigmented or Vascular Skin Marks (RPPVSM): Skin-Based Personal Identification on Digital Evidence

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After attending this presentation, attendees will gain understanding on Relatively Permanent Pigmented or Vascular Skin Marks (RPPVSM) for identifying criminals and victims in digital evidence, especially in photographs and videos. This includes uniqueness of RPPVSM patterns, statistical analysis of RPPVSM, potential error rates in matching RPPVSM evidence, and reliability of RPPVSM recognition by dermatology experts. RPPVSM include but are not limited to nevus, lentigo, cherry angioma, and seborrheic keratosis.

This presentation will impact the forensic science community by exposing it to scientific basis and practical application of RPPVSM in forensic investigation. Attendees will be presented with dermatological description of RPPVSM and their stability over time, a statistical model for estimating the discriminative power of RPPVSM patterns, potential error rates in claiming an identity based on RPPVSM patterns, and inter-examiner variability of RPPVSM analysis in digital photographs.

As multimedia technology becomes more prevalent in everyday life, the capability for extracting evidential information from images and videos during forensic investigation becomes increasingly crucial. For example, in the attempt to search for suspects of child sexual exploitation offenses (e.g., child pornography), forensic investigators often link related digital evidence images/videos found in different cases. Not rarely, the collected evidence images/videos show only non-facial skin of the suspects (e.g., back, chest, thigh, and arm), making face recognition impossible. While tattoos, scars, and birthmarks have some information for identification, they are not always available. However, it is not unusual to observe some other skin marks in these images/videos (e.g., nevus), which can provide valuable information to the investigation.

Skin marks have been regularly used in criminal investigation for years. Birthmarks and nevi are commonly utilized in suspect verification. Nevi were used in the trial of *United States v. Michael Joseph Pepe (2008)*, a sexual exploitation case involving seven pre-teen girls in Cambodia. In this case, the pedophile was identified using a nevi pattern located on his left thigh. The nevi were recognized by a U.S. board certified dermatologist. While it had been known that skin marks are useful for identification, unlike in fingerprint and DNA analysis, their scientific basis for identification had not been established at that moment. The number of skin marks sufficient for authenticating an individual, the error rates of such an identification approach, and the reliability of RPPVSM recognition by experts are fundamental questions that require scientific studies to answer.

Over the past three years, significant research efforts have been made to answer these questions. Statistical analysis on Caucasian, Asian, and Latino populations revealed that Caucasians tend to have more RPPVSM than Asians and Latinos, and that RPPVSM tend to form either a clustered distribution or a uniform and independent distribution.¹ Individuality study on the independently and uniformly distributed RPPVSM demonstrated that seven RPPVSM can be considered sufficient for individualization.² The error rates in verification and identification can be estimated using a statistical point-matching model.² Current research effort is focused on the assessment of the reliability of RPPVSM recognition by different experts. Four dermatology-trained medical doctors have been recruited for this study. Each expert will perform RPPVSM recognition on over 50 Caucasian and Asian skin images

with different quality (e.g., compression rate, image contrast, and occlusion). The consistency among different experts will then be analyzed. Furthermore, the effect of different image quality on their performance will also be studied.

This work is partially supported by the Ministry of Education, Singapore through Academic Research Fund Tier 2, MOE2012-T2-1-024.

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1. Nurhudatiana, A., Kong, A.W.K., Matinpour, K., Cho, S.Y., and Craft, N., "Fundamental Statistics of Relatively Permanent Pigmented or Vascular Skin Marks for Criminal and Victim Identification," in *Proc. Int. Joint Conf. Biometrics*: 1-6, 2011.
2. Nurhudatiana, A., Kong, A.W.K., Matinpour, K., Chon, D., Altieri, L., Cho, S.Y., and Craft, N., "The Individuality of Relatively Permanent Pigmented or Vascular Skin Marks (RPPVSM) in Independently and Uniformly Distributed Patterns," *IEEE Trans. Information Forensics and Security*, 8(6): 998-1012, 2013.

Skin Mark, Identification, Digital Evidence

B18 Forensic Hand Comparison

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After attending this presentation, attendees will learn about issues with hand comparison and the use of digit ratios in forensic images.

This presentation will impact the forensic science community by illustrating how, in child sexual abuse cases, hand comparison can be useful for finding a relationship with the perpetrator. The limitations and possibilities are shown also in databases of hands of siblings and other public-available databases.

In certain cases, such as child pornography, the faces of the perpetrators are not visible in the images. This makes it hard for forensic investigators to find the perpetrator. However, the hands of the perpetrators, are sometimes present in the image. This research investigates different characteristics of hands in order to find certain hand characteristics which in the future might be used to help identify perpetrators.

In the first test performed, all of the finger length ratios from the Bogazici University database in Turkey have been calculated. The ratios of D2:D5, D3:D5 and D4:D5 are found to have the widest dispersion. These ratios might in the future, when more research is performed, be used to calculate a likelihood ratio.

In the second test, left and right hands of these Turkish individuals were compared to investigate the extent to which these hands are similar. Between the left and right hands the digit ratios differed the most. However, there was almost no difference between the length of the fingers of the left and right hand.

For the third test a database of Dutch siblings was built. A total of 80 siblings (31 men and 49 women) participated in this research.

As a negative control, random individuals were compared to each other. For this test the first individual of every family was compared to the first individual of the subsequent family. This way there was data generated, using the same methods as was used for the siblings, when comparing non-family members. The data of the sibling's investigation is also divided for men and women to see if there are specific characteristics more abundant in either men or women.

In the fourth test five face comparison experts from the Netherlands Forensic Institute were given the questionnaire used in this research and asked to compare five sets of hands. These images of the hand pairs were images of the palm of the hand. The experts were not given a positive and negative control. The different characteristics which were present on the questionnaire could be classified as being 'similar', 'different' or 'non observable'. It has been statistically tested if these researchers had different results from the comparisons using this questionnaire compared to the researcher who conducted all

The most distinctive characters between siblings were the birthmarks and scars on the back of the hand. The shape of the hand was found to be a good indicator for kinship. Also, freckle patterns are found to be more common on female hands than on male hands and the hand palms of brothers differ more than the hand palms of sisters. This research shows that in the future, when more research is performed, digit ratios can be used for forensic comparison.

Hand Comparison, Biometric, Internet

B19 New Methods for Linking Cameras on Social Media

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The goal of this presentation is to inform attendees about the possibilities and limitations of linking user profiles on social media to each other via images containing Photo Response Non-Uniformity (PRNU) patterns. An easy way to speed up the process of camera identification by using greyscale images will also be shown.

This presentation will impact the forensic science community by showing the effect that compression added to images by social media databases has on the ability to identify a common source of these images and by demonstrating a simple and effective way to decrease computation time by using grey scale images.

Many social media websites allow for the uploading of digital images. Sites like Facebook®, Flickr, Twitter, and Photobucket contain large numbers of images. Aside from their intended use, these sites are also used for illegal activities, such as spreading child pornography, scamming, fraud and terrorism. Generally these activities are done anonymously.

Using PRNU patterns images can be compared to each other to find out if they have a common source. Via this method a link can be established between an anonymous account used for illegal purposes and a normal user account on a social media website.

When an image is acquired with a digital camera light is captured with a camera sensor. Pixels on the sensor all have a slightly different sensitivity to light. This adds a systematic noise to an image which is approximately the same in each image taken with the same camera sensor. This noise is called the Photo Response Non-Uniformity and is caused by imperfections during the manufacturing process. By extracting and comparing the PRNU pattern from images it can be determined if they have a common source.

By comparing the PRNU pattern from images found on the social media page of a known user to the PRNU pattern from

images found on the social media page of an anonymous user a link can be established. This can help identifying possible suspects.

Uploading an image to a social media website can severely reduce image quality by adding a layer of JPEG compression and resizing the image dimensions. During this presentation the possibilities and problems of determining a common source of images uploaded to these websites will be discussed.

Some sites add more compression than others. Furthermore, it is dependent on the settings used, image quality of the uploaded image before compression, and the camera. However, after such heavy compression it is often still possible to determine if images come from the same camera; even a comparison of two single images can still work.

Most images consist of three color layers: red, green and blue (RGB). Before extracting the PRNU pattern these layers have to be separated. This results in three layers with the same size as the original image. If the image was first converted to greyscale only one layer would have to be extracted, reducing calculation times and pattern size by a factor of three.

While this is not a new concept, as far as we know no data has been published to show the effects of converting to greyscale. During this presentation data of several RGB-to-greyscale ratios and their impact on camera identification will be discussed. It will be shown that using images converted to greyscale is at least as effective as using full color data.

PRNU, Social Media, Big Data

B20 Image Attribution in Social Media

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After attending this presentation, attendees will understand how to determine the source of images posted to social media.

This presentation will impact the forensic science community by providing a tool for law enforcement to track social media images posted to Facebook®, MySpace®, and Google Plus® and methods for applying the same comparisons will any social media website. This presentation will seek to eliminate the mystery that has surrounded images uploaded to the internet.

Society has embraced social media that is rapidly becoming interwoven in our daily lives. Traditional eye witnesses of crimes are being replaced by video and image recordings. Today, almost everyone owns a cellphone and crimes are being increasingly captured on cellphone cameras. Unlike the traditional camera, people always carry their cellphones and they are becoming increasingly user friendly. Due to proliferation and ease of use, cellphones are becoming increasingly important to law enforcement investigations. The largest social media website, Facebook, reports 1.11 billion active users and 300 million picture uploads every day. The combination of cellphone camera technology and Social Media is making it easier than ever to capture images and publish them on the internet.

Investigators now have to determine how to deal with pictures posted to these social media websites that become evidence in crimes. Traditional methods of authenticating the origin of pictures are useless because the processing applied by the social media websites eliminates or obscures much of this information. This presentation will cover several methods of image attribution utilizing both simple data analysis as well as more advanced statistical comparison. This presentation will provide the novice with simple tools and the expert with more statistically precise tools to determine the source of pictures found in social media.

Image Attribution is the process of determining the origin or processing path that an image has gone through. Three Image

Attribution methods will be discussed: Social Media Website Image Attribution, Camera Model Image Attribution, and Specific Image Attribution. Social Media Website Image Attribution determines which social media website processed an image, Camera Model Image Attribution determines which camera model an image originated from, and Specific Camera Image Attribution determines which specific camera an image originated from.

An image corpus was created to test the each of the three methods. 1,100 images were taken with 9 separate cellphone cameras and uploaded to three social networking websites. 1000 images from each camera were used to determine an image database and the remaining 100 images were used for comparison. This corpus will determine how accurate each method by determining Error Rates and Likelihood Ratios.

A combination of simple data analysis and Image Authentication techniques were used to determine signatures in the test images and match the comparison images to the database images. Simple data analysis methods include metadata analysis, structure analysis, and quantization table analysis. Image Authentication techniques used included Compression, Color Filter Array, and PRNU analysis.

The final results of this research prove that Image Attribution is possible for determining the social media website, camera model, and specific camera with a high degree of certainty.

Image Attribution, Image Authentication, Social Media

B21 An Efficient CCTV Camera Calibration Method With an LED Ruler for Body Height Measurement

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After attending this presentation, attendees will understand some principles of height measurement and efficient calibration techniques for Closed Circuit Television (CCTV) cameras.

This presentation will impact the forensic science community by providing a simple method compared with a traditional method using chessboard. A proposed method is useful for crime investigators who pay special attention to morphological characteristics of suspects or subjects in photograms taken from CCTV cameras.

When investing a crime, images taken from CCTV cameras are submitted as clues of investigation. Body height estimation for a suspect or suspicious person in the images is often requested. Camera calibration is a crucial step to extract morphological information from 2D images and much work for camera calibration has been achieved in the photogrammetry and computer vision community. A planar pattern plays an important role in camera calibration methods. The traditional technique for camera calibration with chessboard is usually accomplished by an effective algorithm based on automatic recognition and matching of chessboard pattern.¹ Camera calibration, however, using chessboard is not practical for wide-range images taken in premises of a station or a department store. In this report, a novel camera calibration method with 1D objects using a LED ruler is proposed. Camera calibration method with 1D objects was proposed by Zhang and the 1D objects (stick) was fixed at one point.² A LED ruler which stand perpendicular, that is, fixed at a vanishing point is used as 1D objects in a proposed method in this paper. 3W white LEDs put into eight places (200cm, 180cm, 160cm, 140cm,

120cm, 100cm, 60cm, and 5cm) on the scale of the ruler. The LED ruler move up and down automatically and is adjusted to ensure a vertical position. Calibration points are recorded at each places in a CCTV image. The coordinates of calibration points is used for vanishing point estimations and calibration of the lens distortion. For body height measurement, a virtual LED ruler interpolated by adjacent calibration points is provided on foot point. A prototype camera calibration system with a LED ruler was developed that successfully measure a suspect height. Attention is drawn to the uncertainty in body height measurements and the limitations of the number of the calibration points in an image. Results of testing the prototype in a laboratory setting could measure a height with a difference of less than 1cm. As the number of calibration points in an image decrease, the accuracy of height estimation become worse. Using nine calibration points in an image photographed above the specified size at 640*480 pixel resolution for 15m*20m region, Root Mean Square (RMS) for the height difference was 3.1cm. This is not significantly less, but the accuracy of body height measurements with cross-ratio method has been improved.³ The proposed calibration method using a LED ruler for body height measurement improves the speed of calibration work in the field.

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3. A. Criminisi, I. Reid and A. Zisserman "Single View Metrology" International Journal of Computer Vision 40(2),123-148, 2000

Height Measurement, Camera Calibration, Photogrammetry

B22 Quantitative Comparison of Frame Extraction Methods for Motion JPEG Video

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After attending this presentation, attendees will understand some principles in digital image analysis, digital video processing, and quantitative analysis.

This presentation will impact the forensic science community by providing various methods for the quantitative comparison of frames that should be identical or as close to identical as possible. These methods can be used in a forensic lab when evaluating software or processing options during the development of Standard Operating Procedures (SOPs).

The fundamental principle of forensic science and in the handling of digital and multimedia evidence is to ensure the integrity of evidence through the collection and analysis stages of an investigation. Safeguards such as write-blocking media and hashing copied data are integral to this principle. However, it is universally understood that this is not always possible, such as in the lifting of fingerprints or with the live analysis of a computer. When circumstances arise which require a change to evidence, those changes should be necessary, understood, documented, and explainable in a report or to a jury.

In the processing and analysis of forensic imagery, changes to evidence are common in order to clarify details that may be obscured due to poor lighting, compression artifacts, system resolution, etc. Digital enhancement or clarification of imagery has long been acceptable in court so long as the processing steps taken are documented and the original imagery is available. However,

minute changes to digital imagery can occur during the extraction and preparation of material that are often times unavoidable, albeit unnoticeable, such as in the common procedure of extracting video frames uncompressed from compressed video sources. This study will present the results of a quantitative comparison of extracted uncompressed frames from a motion JPEG video where a ground truth video frame is available. In this particular situation, the minute changes derive from implementation of the DCT decoding algorithm necessarily employed to process JPEG images. The ground truth JPEG frame, available by carving its data from the JPEG video stream, represents unchanged evidence and can be compared pixel-to-pixel to frames extracted through various means. This shows which method will change the evidence frame the least; an important consideration not only when respecting the aforementioned fundamental principle but also since it is necessary to maintain as much original detail as possible when analyzing and interpreting evidence.

Two methods will be demonstrated for quantitative evaluation of extracted frames where a ground truth is known. The first will use Photoshop® and probably be a more comfortable approach for image analysts while the second approach will employ MATLAB to derive more thorough statistical information for evaluation.

Media Forensics, Image Analysis, Forensic Video Analysis

B23 Standardizing 3D-3D Facial Superimposition for Identification From Next Generation Video Surveillance Systems: A New Challenge for Forensic Anthropology and Digital Image Experts

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After attending this presentation, attendees will gain knowledge concerning advanced technologies and the modern applications of forensic anthropology in personal identification from video surveillance systems of living individuals.

This presentation will impact the forensic science community by providing a new method for assessing identification of culprits filmed by 3D video surveillance systems.

The identification of culprits from video surveillance systems is frequently performed by comparing facial features and presents relevant problems regarding both the reliability of methods used and the quantification of the degree of matching between the culprit and the suspect's face. Video surveillance devices provide a film of the scene, and therefore give only 2D images of the culprit, which can be compared to a 3D model of the suspect's face, usually obtained by a laser scanner or stereophotogrammetry, in a 2D-3D superimposition. The attempts actually recorded in literature at standardizing and quantifying the probability of identification are applicable only to ideal conditions of head position and quality of image, and do not provide a conclusive indication concerning the identification.

However, in the near future video surveillance systems will provide a 3D image of the filmed person, which may allow the operator to perform a 3D-3D superimposition between the 3D model of culprit's face and the 3D scan of the suspect. The application of 3D-3D superimposition promises to overcome the limits concerning the probability of correct identification, a difficult

task in 2D-3D comparison techniques.

A pilot study on 55 3D-3D superimpositions was devised: the face of ten individuals underwent two scans by stereophotogrammetry (VECTRA-3D) which provided a 3D model of the face of each subject. The time period elapsed between the two scans went between few minutes and 32 months. On each 3D facial model, nine landmarks (right and left endocanthion, right and left exocanthion, right and left cheilion, on the midline selion, pronasale, and subnasale) were identified by VAM software; the first scan of each individual was then superimposed with the first scan of all the other nine subjects. The superimposition of the two 3D models was performed in order to reach the best match between the corresponding landmarks. The same procedure was performed also between the two scans taken from the same individual. In total, 55 superimpositions were performed. In all the cases, the Root Mean Square (RMS) value of point to point distances between the two models was calculated. When the two superimposed models belong to the same individual, the RMS value was on average 1.58 mm (SD: 0.61 mm), whereas when the two models were taken from different persons, the same value was 4.54 mm (SD: 2.02 mm). The difference between the results obtained in the two groups was statistically significant ($p < 0.0001$).

This pilot study shows that the judgment of identification based on 3D-3D superimposition may provide reliable results concerning the definition of a threshold for reaching a diagnosis of identification; in addition, this protocol may allow scientists to give a probability concerning identity. The improvement of technologic devices of scene acquisition and the development of specific methods of 3D-3D comparison may provide in the near future relevant advantages in this recent and difficult field of forensic anthropology.

Personal Identification, Video Surveillance System, Stereophotogrammetry

B24 Why Automated Face Recognition Cannot Be Used to Eliminate Suspects

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After attending this presentation, attendees will understand that: (1) automatic Face Recognition (FR) is not designed to eliminate suspects, but to find potential matches; (2) even the best FR algorithms will have high false negative rates under a variety of conditions; and, (3) the variety of factors that lead to false negatives is so great, that using FR to eliminate suspects in the future is only likely to occur under the most restrictive of conditions.

This presentation will impact the forensic science community by making them aware of a key limitation of face recognition technology. It will also remove the presumption that just because a technology may be useful at including a subject in a pool of suspects, it cannot be assumed that it is equally capable of eliminating a subject.

Automated facial recognition (FR) has become an extremely useful tool for law enforcement and related government entities. For example, face recognition technology now allows numerous states to combat identity fraud by preventing criminals from obtaining driver's licenses under multiple identities. It was recently reported that New York State has had over 2,000 arrests for fraud since 2010 thanks to FR.¹ Likewise, law enforcement agencies have started to use FR to locate criminals who left their pictures behind while committing crimes.²

The success of these efforts results from over twenty years of steady improvement in the algorithms used to match faces. The most recent large-scale tests published by the National

Institute of Standards and Technology (NIST) documented that the best face recognition algorithm tested had a false non-match rate of 0.3% when measured against a fixed false match rate of 0.1%.³ This is a remarkable improvement, especially when one considers that the false non-match rate was close to 80% in 1993 when the NIST tests began!³

The success of FR has led some in the criminal justice system to assume that it can be used equally well to exonerate individuals. This is not the case. While the NIST results provide strong evidence that FR can be used to identify potential matches, these tests were not designed to assess the degree to which FR can be used to eliminate subjects. Indeed, the point of this is made: "...a low score does not necessarily mean the images are of different persons. This arises because defective images produce low scores even in same-person comparisons. The term defective might mean low contrast, blurred, non-frontal pose, and exaggerated expression."³ Such "defects" are likely when comparing controlled images (e.g., mug shots) against uncontrolled images acquired from sources such as closed-circuit television (CCTV) systems. Likewise, other factors, such as aging and illumination changes, can also lead to low scores (i.e., false non-matches).³

In this study, a Government-OTS FR algorithm is used to demonstrate the variation in match scores for a number of same-person comparisons involving controlled and semi-controlled images. Although some high match scores are correctly returned, in many cases low scores are returned, with many falling below match scores generated in different-person comparisons. These results demonstrate the inability of current FR approaches to serve as a reliable means of eliminating suspects under all but the most highly controlled circumstances.

Given the variety of factors that can lead to low-match scores in same-person comparisons, it will be a challenge to acquire controlled datasets with sufficient sample size and variability to address all of these factors for all relevant populations. Instead, the best chance for developing FR algorithms to support the elimination of subjects will probably be limited to highly controlled applications.

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Face Recognition, Biometrics, False Negative

B25 Analysis of Manipulated Photographs Submitted to Federal Court and the United States Congress

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After attending this presentation, the attendees will have

an understanding of the features of a manipulated photograph submitted to federal court that was likely fabricated in 1993 or early 1994 as well as another manipulated photograph that was submitted in a report to the United States Congress in 2004.

This presentation will impact the forensic community by providing investigative techniques for the detection of manipulated photographic evidence.

Images are routinely recorded in many autopsies, and may be useful in ascertaining the details of the cause of death. The analysis and interpretation of those images generally assumes that they are properly taken and handled, and can be relied upon. This paper concerns a case in which manipulation of such an image may have led to an unfounded conclusion.

In 1994, as a result of a wrongful death civil case in federal court (*Sara Sabow et al. v. The United States, US District Court, Central District of California, Southern Division. SA CV 933-991 AHS*), the defendant, the United States Department of Defense, released to the plaintiffs approximately 30 autopsy photographs of the decedent. The decedent's autopsy was performed by an Orange County (California) Coroner's medical examiner. The plaintiffs alleged the decedent died due to a powerful blow to his posterior head on the right occipital and not by an intraoral shotgun blast which followed the blow to the head. The defense claimed the victim died solely by the intraoral shotgun discharge and the death was by suicide.

One of the autopsy photographs differed from all the other autopsy photographs by being poorly taken. This photograph of the victim's posterior upper body appeared to have had the flash too close to the victim's lower left side, which caused the upper back, neck, and left pinna to be overexposed. Most of the head and background was cast into deep shadow. Simulation of the lighting conditions on a mannequin produced a similar image, but the apparent lack of illumination of the victim's head in the questioned photograph could not be duplicated.

Digital analysis of a high resolution image of the questioned photograph shows that it was likely the result of manipulation, either in the darkroom or, less likely, image processing. It was determined that the questioned photograph, unlike the other autopsy photographs, lacked background detail within its deep shadow. The deep shadow areas also had fine parallel stripes when lightened by increasing gamma. In lighter regions of the photograph, parallel stripes were also apparent and perpendicular to the stripes discovered in the deep shadow areas. Similar stripes have been described by Russ as a common defect of flatbed scanners of this period.¹

It was determined that a nearly identical photograph to the questioned photograph that was submitted in a report to the US Congress in 2004 was a different fabrication. Thus, there are two photographs submitted in this case that have indications of improper manipulation.

In December 2012, the original unmodified photograph was discovered from which both manipulated photographs were apparently generated. The original photograph allowed for a more detailed analysis of the questioned photographs and proved beyond reasonable doubt that perjured submissions had occurred: the first in federal court and the second to the United States Congress.

The original, unmodified photograph shows the purpose of both image alterations was to hide the pronounced swelling of the victim's upper right neck which is attributed to a premortem blow to the back of the victim's head. This suggests that there was knowledge of a blow to the head prior to the intraoral shotgun discharge and the decedent was a victim of homicide.

An image of the second altered photograph was extracted from the pdf file of the report to the U.S. Congress. Clipping of the blacks by pdf conversion and low resolution prevents a detailed analysis.

The author of the 2004 report to the U.S. Congress, which

contains the second manipulated photograph, has been informed of these findings and has not responded.² It is strongly suggested that this expert should re-evaluate this new evidence and determine if it would impact his previous conclusions on this case.

Many thanks to Dr. John C. Russ for his assistance in this study.

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Manipulated Photograph, Imaging Processing, Federal Court

B26 Legal Processes for Cloud Forensic Investigations

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After attending this presentation, attendees will gain an understanding of the legal challenges involved in cloud computing investigations and the potential legal processes that may be used to demonstrate the admissibility of cloud-based forensic evidence.

This presentation will impact the forensic science community by providing a legal perspective on the emerging area of cloud forensics. Attendees will learn what steps may be necessary to authenticate potential forensic evidence from public cloud computing providers for criminal or civil investigations.

Organizations have continued to adopt cloud computing as a cost-effective processing and storage platform. As the cloud's prevalence increases, it will have an increasing impact in legal proceedings. Once cloud-based data is obtained, it is minimally useful unless it is accepted as evidence in a case. The Federal Rules of Evidence (or similar applicable rules) were originally designed for paper documents, and updates to address digital evidence have yet to consider the cloud's different paradigm. Unlike traditional hard drive forensics, forensics involving cloud computing does not include the ability retain a physical, original hard drive. The lack of original, physical-based forensic evidence presents admissibility and authenticity concerns.

Authenticity concerns are the main barrier for the use of cloud-based data in legal proceedings.¹ Cloud-based data that is obtained from a public cloud is, by definition, physically comingled. When challenged in legal proceedings, a party must be able to demonstrate that the data produced is associated with a particular user. While this may be established through methods including testimony of a witness with knowledge, additional concerns arise when the admissibility challenge is targeted toward the actual processes and functions of the cloud computing environment. When the actual process for obtaining cloud-based evidence from a particular cloud user is challenged, a party may be required to prove the reliability of the underlying cloud computing process or system. This may require expert witness testimony from the cloud provider.

A walkthrough will be provided of the applicable rules of evidence that may apply to cloud-based forensic data. The potential processes for admitting and authenticating cloud-based evidence into a case will be explained. These potential processes have been modeled to address the particular unique issues of cloud-based evidence. Other hurdles that may arise when authenticating cloud-based evidence including the Daubert test for digital evidence will be discussed.

In conclusion, the nature of cloud-based forensic

evidence requires new applications of legal processes. This presentation will provide the legal and digital forensic communities with a fundamental understanding of such processes, and potential challenges for the use of cloud based data.

Reference:

1. See I. Orton, A. Alva, and B. Endicott-Popovsky, "Legal Process and Requirements for Cloud Forensic Investigations," *Cybercrime and Cloud Forensics: Applications for Investigation Processes*, IGI Global, 2013, pp.186-235; <http://ssrn.com/abstract=2197978>

Cloud Computing, Digital Forensics, Legal Processes

B27 Google™ Dual-Factor Authentication (2-Step Verification) and Its Impact on Cloud Data Collections

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After attending this presentation, attendees will understand the recent move toward dual-factor authentication by online service providers such as Google™ to offer increased data/information security to its users, and the impact this can have on forensic data collections carried out in connection with litigation (which are often done by 3rd parties such as law enforcement officers or private practitioners hired to accomplish the same). Attendees will learn, as an example, how Google™ implements this technology, and what they will need to do in order to accomplish forensic data collections from Google™ when necessary for either civil or criminal litigation.

This presentation will impact the forensic science community by introducing this new and lesser-known technology, explaining various aspects of its implementation, and providing methods to properly address it so that forensic data collections can take place more quickly and smoothly than would otherwise be the case.

Recent security changes implemented by online data service providers such as Google™ have resulted in some changes to the way information must be accessed, collected and verified from Gmail accounts, and mail accounts hosted through Google™ Apps (<http://www.google.com/enterprise/apps/business/>). In order to avoid collection delays and frustrations, law enforcement officers and companies should prepare themselves ahead of time for dealing with these issues during a collection.

There are two relatively new security measures implemented by Google™, and accompanying best practices, that are important to be familiar with. The first is the use of Verification Codes.

In order to increase security, Google™ sometimes requests a verification code when a user attempts to access a Gmail account from a browser or machine that has not previously accessed an account. Without the verification code, the account cannot be accessed. Verification codes can only be obtained by the account holder in certain ways (e.g., a text message sent by Google™ to a pre-configured cell phone; via an application called "Google™ Authenticator," etc.).

Part of the collection process typically involves logging into the email account through a browser to verify message counts and other information, which may prompt a Google™ request for a verification code. To speed collection times, it is important to: (1) be aware of any users with mobile devices linked to such cloud accounts for the purpose of retrieving or generating verification codes; (2) provide the collections team with access to these users and their mobile devices as necessary; and, (3) alert these users

that a collection will be occurring, and ask them to be ready to provide verification codes during the collection. Valid verification codes change every 30 seconds, so collection teams must work with these users in real time.

The second relatively new security measure implemented by Google is the use of 2-step verification. If a user or organization has chosen to enable 2-step verification for their Gmail or Google Apps accounts, collection teams will both need to obtain a verification code as noted above, and obtain an "application specific password" for any tool (e.g., Microsoft® Outlook) used to access the account that does not do so through the use of a browser. The application specific password can only be generated by the custodian or the account administrator. To speed collections when two-step verification is enabled, it is important to: (1) allay privacy concerns with custodians before the collection begins; and, (2) prepare custodians or account administrators for the process that generating the application specific passwords will take.

Collecting data from the cloud isn't as easy as it may seem to many attorneys, judges and others involved in litigation who may make such requests. Enhanced security features like those implemented by Google have the potential to delay forensic collections. With a little preparation however, law enforcement officers, legal and IT teams can ease administrative headaches and overcome obstacles to a successful collection.

Google™, Authentication, Data Collection

B28 Acquisition Issues With Cloud Computing

Ernesto F. Rojas, MBA, PO Box 597, Seabrook, TX 77586*

After attending this presentation, attendees will be able to understand the principal issues that exist in operating in a public cloud computing environment. Examples of problems with multiple providers, data recovery, electronic discovery document collection, and other areas of interest will be addressed while contrast and comparison to traditional computing environments will also be discussed.

This presentation will impact the forensic science community by addressing issues related to the acquisition and migration to cloud computing environments by commercial and government entities and how they affect regulatory and legal requirements.

Cloud Computing has for the past two years become the latest must have of the ever-exploding computing innovation world.¹ Computing as a utility has many features that make the transition to the services extremely attractive by transferring capital requirements and technical responsibilities for operations along with other attractive features. In the process of rushing to order the latest cloud based software or service the user community is ignoring many important issues that will, once adopted, come to bear unforeseen problems at the worst possible moment.²

One of the principal issues in using cloud computing is the location of the data. A review of cloud computing service agreements show that the majority do not address the location of the data, leaving physical and jurisdictional locations to the decision of the cloud provider. This issue of location becomes important when there are legal requirements to keep certain type of information in conformance with regulatory and judicial regulations.³

Security is another major issue that in many cases goes unmentioned in cloud computing agreements; this is a major problem with Application-as-a-Service providers where the provider is asking the consumer to fully trust that the provider is handling all security issues to prevent theft and/or unauthorized manipulation of the users data.⁴ Security in the cloud is rapidly becoming a world of specialists that provide security and authentication services

to Cloud Service Providers (CSP) as a service, separating the operating and legal responsibilities and allowing for the transferring of risks to users in making sound choices in the selection of services.^{5,6} In summary, this presentation raises issues that need to be considered before committing to a Cloud computing environment by a prospective user.

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Cloud, Business, Computing

B29 Mapping the Forensic Standard ISO/IEC 27037 to Cloud Computing

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After attending this presentation, attendees will be able to summarize the current integration of cloud forensic requirements into service level agreements (SLAs), and understand the international standards for cloud forensics. In particular, attendees will be able to understand how the Cloud Security Alliance mapped cloud computing to forensic standard ISO 27037 ("Guidelines for identification, collection, acquisition, and preservation of digital evidence").

Since the forensic process for cloud computing largely hinges on the legal contract between customers and providers, this presentation will impact the forensic science community by illustrating the Service Level Objects (SLO) that must be incorporated into service level agreements between customers and Cloud Service Providers (CSP).

In the short-term, the cloud consumer bears the responsibility to ensure that CSPs can respond appropriately to a forensic investigation. Consumers ultimately suffer the loss from crimes in the cloud environment. When contracting for cloud services, the customer should ensure that explicit language and SLOs are incorporated into the contract to ensure they can respond appropriately when the need to perform a digital investigation arises. For CSPs, integrating forensic capabilities into cloud offerings would increase transparency for the consumer and likely lead to greater revenue streams. As more organizations become reliant on cloud computing for critical operations, we foresee that forensics will become a key motivator on choice of CSP. Additionally, as the cloud market matures, legal and regulatory changes are foreseen that may shift duties to include, collaboratively, CSPs.

ISO 27037 is an international standard that seeks to create a baseline for the practice of digital forensics.¹ Not intended to usurp local or national governmental authority, the standard's intent is to facilitate the usability of evidence obtained in one jurisdiction by a legal process operating in another jurisdiction. In its present form, ISO 27037 addresses identifying, obtaining and preserving

potential digital evidence.

ISO 27037 is a relatively new standard (issued in October 2012) and only addresses the initial stages of a digital investigation, but it represents an international public and private sector consensus of how potential digital evidence should be handled in the critical initial steps of an investigation. There are many complex challenges of digital forensics in a cloud environment and how CSA mapped and reinterpreted the ISO 27037 guidance for a cloud context is explained.² For some parts of the standard, no changes are necessary for cloud environments. For others, including identification and acquisition of evidence, cloud requires special considerations.

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Digital Forensics, Cloud Computing, Forensic Standards

B30 A Proposed Cloud Computing Forensic Interface

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After attending this presentation, attendees will be able to understand the need for a common interface that enables forensic practitioners to extract data, metadata, and other log files and to conduct an investigation of the events related to a particular case. This presentation is a proposal to the forensic cloud computing community to consider development of a common interface to facilitate training, consistency of results, and commonality of forensic collections among multiple cloud providers.

This presentation will impact the forensic science community by encouraging discourse among members of the forensic cloud computing community to recognize the difficulty that investigators have in learning multiple forensic interfaces from one provider to another and the time savings created by a common user interface for forensic collections.

At the present time, Cloud Computing providers have either developed a method or interface for the collection of evidence from their cloud environment and in many cases there is no interface provided to the user community for evidence collection. This disarray is rapidly becoming a major obstacle for cloud users, law enforcement, the legal community, the courts, and government agencies to collect information to support an investigation, when cloud applications and storage are employed as part of the computing environment storing data related to a legal dispute or incident. This presentation proposes a model that has a common user interface to effect the collection of evidence, with the back-end of the model customized to interface with the provider's existing software architecture, so that when files are collected they are presented in a forensically sound format, with their metadata intact in a similar manner as files collected by current digital forensic processes available today.

This proposal is based on foundational work done by Dykstra and Sherman for the FROST interface developed and the experience in forensics of the presenter.¹ A list of items that should be part of the interface will be offered as a starting point from

which to develop the menu for the interface, along with a proposed method by which to have a back end that will be easy to connect to the cloud providers existing software architecture. In addition the use of existing forensic evidence formats will be suggested as a way of standardizing the output of the interface.² In conclusion, this presentation proposes to stimulate the conversation to adopt a common format for the collection of evidence from cloud providers.

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Cloud, Forensic, Interface

B31 Credentialing the Digital and Multimedia Forensics Professional

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After attending this presentation, attendees will have gained an understanding of credentialing and its application to digital and multimedia forensic science as a profession, as well as the status of current credentialing efforts as they are evolving for digital and multimedia forensics.

This presentation will impact the forensic science community by providing criteria for assessing vendor-neutral credentials and examining several contenders for credentialing digital and multimedia forensics professionals.

Professions are characterized by some degree of monopoly rights to practice based on formal qualifications supervised by regulatory bodies with power to admit and discipline members of the profession.¹ Professionals enjoy high status and prestige.² To be recognized as a professional, one has to acquire specialized knowledge through education, training, and experience, which has to be maintained through continuing education. Admissions are based on some combination of education, experience, and ability to pass an examination, while discipline is based on a code of ethics recognized as binding on the profession.

In the Anglo-American legal system, forensic science grew out of the need for juries to understand scientific, technical or other specialized evidence and its implications for resolution of legal issues.³ The classical professions were medicine, law and divinity, and, to some extent, later evolved professions are modeled on them. Other occupations became professions in the United States, including surveying, actuarial science, dentistry, civil engineering, logistics, and accounting.⁴ By 1900, architects, pharmacists, veterinarians, nurses, teachers, librarians, optometrists and social workers could claim to be professionals.⁵

Digital and multimedia forensics is the newest entry into the forensic science profession and is currently developing credentials and processes for awarding and recognizing those credentials and disciplining their abuse. Courses and programs in digital and multimedia forensics are currently available or being developed at many colleges and universities. Some States are requiring digital and multimedia investigators to hold private investigator licenses. Some vendor-specific or tool-specific certifications are available.

Of particular interest are the development of vendor-neutral certifications that assess breadth of knowledge and depth of experience, and which require adherence to a code of ethics and

continuing education. The presentation will compare and contrast the GIAC Certified Forensic Analyst certification offered by SANS, Computer Hacking Forensic Investigator certification offered by the EC-Council, Certified Forensic Computer Examiner certification offered by the International Association of Computer Investigative Specialists, Certified Computer Examiner certification offered by the International Society of Forensic Computer Examiners, Digital Forensic Certified Practitioner certification offered by the Digital Forensics Certification Board, and the newest entry, Cyber Forensics Professional Certification offered by (ISC)^{2TM}.

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Digital & Multimedia Forensics, Professionalization, Certification

B32 Assessing the Relationship Between Asperger Syndrome, Hacking, Identity Theft, Virus Writing, and Cyberbullying

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After attending this presentation, attendees will have a better understanding of the relationship between Asperger syndrome and computer deviance, specifically hacking, virus writing, cyberbullying, and identity theft.

This presentation will impact the forensic science community by filling a gap in the literature on the autistic-like traits associated with different forms of computer deviance.

Anecdotal cases suggest a potentially unique relationship between Asperger syndrome (AS) and computer deviance, particularly hacking.¹⁻⁵ However, few empirical research studies have attempted to assess this Hacker-AS link. In addition, previous research does not distinguish between individuals engaged in different forms of computer deviance (e.g., identity thief vs. virus writer), and they have not included crimes where the computer was used as the "tool," such as cyberbullying.

Three primary objectives were the focus of the current study. First, was to determine whether there were differences in the autistic-like behaviors (poor social skills, poor imagination, poor communication, poor attention-switching, and exceptional attention to detail) exhibited by computer deviants and non-computer deviants. For example, are computer hackers more likely to exhibit autistic-like behaviors when compared to non-computer hackers? Second, was to include other types of computer deviant

behavior, such as identity theft, cyberbullying, and virus writing, when assessing autistic-like behaviors. Finally, the third objective was to determine whether autistic-like behaviors distinguished between computer deviants. For instance, are there autistic-like traits that differentiate between individuals who engage in one type of computer deviance (hacking) versus individuals who engage in all computer deviant behaviors (hacking, identity theft, cyberbullying, and virus writing)?

An online survey was completed by 269 university students, which measured the prevalence of computer deviance (i.e., hacking, virus writing, cyberbullying, and identity theft) and autistic-like traits (i.e., poor social skills, poor attention-switching, poor imagination, poor communication, and exceptional attention to detail). 57% ($n = 170$) of students reported engaging in hacking behaviors, 13% ($n = 38$) reported engaging in identity theft, 23% ($n = 66$) reported engaging in cyberbullying, and 8% ($n = 23$) engaged in virus writing. Only two students (.01%) met the threshold for clinical levels of autistic traits according to the Autism Quotient, and both of these individuals engaged in computer deviant behavior.⁶ In contrast to the expectations, the non-hackers actually exhibited more abnormal autistic-like behaviors, specifically a strong attention to detail, compared to the self-reported hackers in the current study. The identity thieves and virus writers exhibited more autistic traits compared to their non-deviant counterparts, specifically poor social skills, poor communication, and poor imagination. On average, cyberbullies reported more autistic traits than non-cyberbullies, specifically poor communication.

Finally, the current study supported the expectation of significant group differences on autistic traits based on the individual's level of computer deviance. Specifically, those individuals who engaged in hacking, identity theft, cyberbullying, and virus writing exhibited more autistic traits, including poor social skills, poor communication, and poor imagination, compared to all other individuals engaging in computer deviant behaviors (1 type, 2 types, and 3 types). However, there were no autistic traits, which discriminated between the other levels of computer deviance (i.e., discriminating 1 type from 2 types). Overall, autistic-like traits were related to identity theft, virus writing, and cyberbullying; however, the overall group mean score for the computer deviants on autistic traits was 20, which is in the intermediate rather than clinically significant (32+) range for Asperger syndrome. Limitations and suggestions for future research are discussed.

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B33 Scenarios for Reliable and Secure Digital Evidence Applying a Hardware Solution

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The goals of this presentation are to disseminate solutions for forensic readiness and to encourage discussion of additional applications for the concept.

This presentation will impact the forensic science community by raising awareness that as the bar is raised on the admissibility of digital evidence due to the successful implementation of the technology described in this presentation, more applications requiring solutions of this type will emerge.

This presentation discusses three distinct scenarios where forensic readiness of devices is accomplished through the application of a hardware solution. The scenarios are: lawful interception of voice communication, automotive black box, and precise farming.¹⁻³ Although these three scenarios come from very distinct and different application domains, they share a common set of security requirements for processes to be documented and data records to be stored that can be addressed with a common hardware solution proposed to in some detail with a focus on the legal admissibility aspects of each.

Traditional approaches to digital forensics are concerned with reconstruction of events within digital systems that often are not built for the creation of evidence; however, many scenarios exist where such devices are expected to produce legally admissible digital data records for which admissibility is relevant.⁴ Technical solutions exist using hardware-based security to bind digital records to a particular state of a device.⁵⁻⁷ This presentation discusses the application of this solution in the context of concrete, practical scenarios along with legal and admissibility implications, and economic feasibility.

The solution is a device that is produced and configured in a way that results in admissible evidence which is correct and reliable as long as the device is not physically manipulated or corrupted. Various types of attacks must be considered in its design, including attacks on communication channels, attacks via various physical interfaces (e.g., USB), or attacks exploiting weaknesses of software running on the device.⁸ If the attacker's goal is to manipulate digital data before it is protected by a digital signature, attacks need to change software or configurations on the device in order to interfere with and manipulate the actual creation of resulting data records. At the heart of the solution is the Trusted Platform Module that binds data records to the status of the device at the time of creation of the data record.⁹ This status can include all executable software started on the device since last reboot, configuration parameters, and other parameters on the device's environment

The presentation reviews the 14 step protocol, that supports successful implementation of a forensic ready device.^{10,11} This protocol has been applied in the three distinct scenarios mentioned above. It was shown, that although requirements for each are quite different, all three scenarios can be realized using the solution proposed in 2012 by the authors. Future work includes identifying and analyzing additional scenarios in which this solution is relevant, and testing the solution in actual circumstances. The

concept of forensic readiness, discussed at length years ago is now realizable and available for specific applications.¹² It is anticipated that as the bar is raised on the admissibility of digital evidence due to the successful implementation of the technology described in more applications requiring this solution will emerge.¹³

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Forensic Readiness, TPM, Admissibility

B34 Digital Forensic Analysis Applied to Real-Time Studies of Social Media in the Cloud in Order to Monitor and Predict Development of Contemporary Political Dissent in Semi-Authoritarian Countries

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The goals of this presentation are to apply digital forensics approaches to a new domain, social media, and to discuss results of preliminary work that sheds light on the patterns of online behavior

of protestors and governments in semi-authoritarian countries.

This presentation will impact the forensic science community by shedding light on activities such as the Arab Spring and uprisings in Russia. A new application of digital forensics principles will also be discussed.

Recent events in different parts of the world have drawn particular attention to the use of Internet-based ICTs within contentious political processes. There is a need for the application of digital forensic techniques, applied to Internet social media, that will assist in the development of models that can anticipate and predict popular uprisings. Social media technologies were widely used in 2011 during a wave of the people's uprisings in the Middle East and North Africa (MENA) region, widely known as the "Arab Spring." During some of these uprisings, the authorities, in addition to their usual practice of heavily censoring the Internet, even resorted to the complete blackout of the Internet and phone communications. But even these last attempts at restricting information and communication access often were in vain. Resourceful dissenters used various techniques to circumvent those limitations and still were able to get access to both the Internet and phone networks. Eventually, these networks played an important role in the success of some of those uprisings.

ICTs-facilitated contentious political events in Russia started with mass anti-Putin meetings in December 2011 in Moscow. Since then, protests in Russia, despite the active and pro-active harsh resistance from the incumbent authorities (including active counter-measures in the Internet domain), continue. Here the Internet plays its unique role as both the main supplier of relevant information and also as a robust communication tool for the dissenters, while the authorities simultaneously conduct their own information game aimed at both counter-propaganda (information war) and suppression of the protesters' informational resources with cyber-attacks of various kind (cyber-war). Knowledge obtained during forensic research of all the respective information flows can help monitor development of the situation in that country and predict next steps which will be undertaken by both sides of the confrontation. Moreover, this knowledge can be of practical use during similar events in other authoritarian states worldwide.

Research into the ICTs-facilitated politically-motivated protest events in semi-authoritarian states has been successful.¹ Further, digital forensics techniques including quantitative methods such as (semi)automatic online textual content gathering and analysis, one-to-one, one-to-many, and many-to-many information exchange, and hyperlink analysis in both hierarchical and network structures, are of great use in mining Russian social media for sentiments and trends which will help better understand and even predict the development of protest events in that country.

This presentation discusses a proposed digital forensics approach that will enable researchers to quickly gather and process information from hundreds of thousands of online accounts in the Russian-language SNS, blogs, and Twitter. Continuous hyperlink analysis of the Russian-language online public space will allow us to continuously monitor its development and transformation in real-time, to enable analysis and prediction of the dynamics of the situation under investigation. Real-time visualization of obtained results will also allow effective monitoring and analysis of the development of patterns surfacing in observed networks of interaction in the Russian Internet domain.

The overarching result of this project will be development of methods and algorithms necessary for the creation of a software/hardware complex able to monitor and visualize in real-time the dynamics of the highly-contentious ICTs-facilitated political events in the non-free countries whose authorities aggressively (counter) act protesters in the online space. This theoretical framework is expected to be extended and will enable classification and dynamic identification of key sources motivating these movements, both

protestors and government provocateurs, as well as the most probable vectors of their expected attacks.

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Social Media, Protestors, Applied Forensics Analysis

B35 Digital Forensics and Service Learning: The Oklahoma Tornadoes Project

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The goal of this presentation is to provide the participants with a case study of using digital forensics in service-learning projects.

This presentation will impact the forensic science community by providing tested examples of using service-learning projects in the digital forensics classroom.

During the afternoons of May 24, 2011, May 20, 2013 and again on May 31, 2013, several large tornadoes touched down in Oklahoma killing numerous people and leaving a path of damage fifty miles long. Not only were lives lost, but computers and other digital devices were severely damaged leaving victims without access to pictures, documents and other vital data stored on these devices. University of Central Oklahoma (UCO), Forensic Science Institute, digital forensics students, using the knowledge and skills they learned and the same high-tech equipment used to solve crimes were able to recover terabytes of treasured memories and important documents for tornado victims. This service learning project provided a rich learning experience for students, integrated meaningful community service, and strengthened ties with the community.

Following these devastating tornadoes, the community pulled together to help tornado victims with their immediate needs of food, clothing and shelter. While those needs were the most urgent and critical following the storms; tornado victims began to take stock of what else they may have lost. Many had computers and other digital devices that stored precious memories in the form of digital pictures and videos that were damaged by the high winds and rain. Others lost important business and personal documents that were stored in digital format. Recovery of this information required specialized assistance and could cost hundreds of dollars. Digital forensics students at the UCO Forensic Science Institute offered to apply the knowledge and skills learned in the classroom to help tornado victims recover important data from their damaged computers.

The National Service-Learning Clearinghouse defines Service Learning as a teaching and learning strategy that integrates meaningful community service with instruction and reflection to enrich the learning experience, teach civic responsibility, and strengthen communities.¹ Service learning represents a potentially powerful form of pedagogy because it provides a means of linking the academic with the practical. There is growing evidence that having students apply theoretical material learned in the traditional classroom in a "real world" setting has a positive effect on student learning and interest in the subject matter.² Service learning projects also benefit the community by providing new perspectives into the work of the university and strengthening relationships with faculty and students.

While all the examinations were not successful, due to the severe damage to the computers and hard drives, students were able to recover several terabytes of data in the form of pictures, videos and documents for 90% of the tornado victims. The ability to return precious memories and vital documents back to the tornado victims gave the students confidence in their abilities as digital forensic examiners and a great sense of pride in providing a service to the community.

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Digital Forensics, Service Learning, Case Study

B36 Collecting Ground-Truth, Web-Based Data for Research in Forensic Linguistics

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After attending this presentation, attendees will: (1) learn about both the utility and the perils of using web-based forensic linguistic research for authorship identification, threat assessment, suicide note assessment, and other language-based issues in criminal and civil investigations; and, (2) be introduced to the Institute for Linguistic Evidence Research (ILER), a web-based platform that addresses the dangers of web-based data collection and provides ground-truth data and automated experimental design for forensic linguistic research.

This presentation will impact the forensic science community by providing document examiners, digital forensic evidence examiners, and other researchers with access to another tool for their work, as ILER provides automated experimental design and text analysis. Because ILER provides ground-truth data, the calculation of accurate error rates is possible.

The world wide web provides an enormous amount of linguistic data, especially through social media sites, blogs, and other fora. It can be extremely tempting for linguists and other forensic examiners to simply “scrape the web” for linguistic data. This procedure has three problems: (1) some ethical issues regarding human subjects;¹ (2) the circularity of using unvetted web-data to solve the problem of anonymous web-data;² and, (3) the procedure does not guarantee ground truth data (necessary for error rates to be correctly and accurately calculated). As more and more linguistic data is generated electronically, the need for collecting data from electronic media is obvious, since the medium of generation may influence the message, harking back to Marshall McLuhan’s communication dictum. Further, as the forensic science community embraces “normal science” and experimental procedures, the need for examining web-based linguistic data is a valid concern, especially when we consider that an experimental paradigm for validation testing requires data collection in a controlled way.³ This presentation presents a platform developed to address these issues, a platform that enables an experimental paradigm in forensic linguistics to use web-generated data in a way that ground truth data can be collected. ILER is a web-based platform that enables researchers and practitioners to design experiments, recruit subjects and collect vetted data via the internet. ILER includes human subject protections to solve the ethical issues. ILER includes automated experimental design so that practitioners can create experiments using their own on ILER

stimuli for the collection of relevant data. ILER is a closed system so that access is monitored, in the same way that laboratory experiments are monitored, so that the identity of the subjects can be monitored as closely as possible, solving the problem of getting ground truth data from the internet. ILER also includes text analysis procedures so that non-linguists can access automated text analysis of the collected data so that the quantification and pattern identification can be analyzed statistically through statistical routines within ILER and from commercial statistical software. Finally, ILER enables alternate means of data aggregation so that the community can share vetted linguistic data for experimental research and validation testing.

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Forensic Linguistics, Authorship Identification, Web Data



Engineering Sciences



SEATTLE 2014

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C1 Forensic Architecture

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The goals of this presentation are: (1) to promulgate awareness of forensic architecture as a forensic discipline; (2) to inform as to the nature of forensic architecture; (3) to show how forensic architecture differs from other forensic sciences; and, (4) to show examples of forensic architecture.

This presentation will impact the forensic science community by demonstrating the usefulness of forensics in the arena of architectural design, design and construction standards, construction, construction law, and personal injury.

This presentation examines three cases being managed to resolution and includes pertinent detail and graphic examples. Management of foreseen actions or events by the forensic architect before an occasion of injury or damage is preventative in nature, such as surveys to identify architectural barriers or participation in development of building design standards. These activities do not become legal situations. However, those arising after the occasion of injury or loss due to non-adherence to building codes or construction specifications usually end up in the legal system.

Case 1: An Americans with Disabilities Act (ADA) study of a county's public buildings and facilities: Sixty county facilities were surveyed for compliance with requirements of ADA and 2,400 non-compliant conditions were found. Facilities consisted of community centers; half-way houses; a hospice facility; senior citizens' and children's day care centers; a cultural affairs facility; a library; a nature center; fire stations and a fire-training academy; water and sewage treatment plants; municipal services and vehicle repair facilities; an employment center; transportation stations; a performing arts center; and a 400,000-square-foot, 12-story detention facility. By working with the legal department of the county, the highest-priority items for remediation were determined at the earliest possible time, making maximum use of the available funds. The consulting contract included expert architectural assistance in legal defense of the county in the event that a complaint is brought by either a person with a disability or by the U.S. Department of Justice. The nature of this forensic architectural service is preventive, providing architectural advice to assist the client before being forced into court.

Case 2: Defense of the owner of air cargo building at any international airport. In the process of selling a 60,000 square-foot air-cargo facility (which included leased space for four large national air-cargo shipping companies), the buyer claimed that the owner/seller had to make over \$500,000 in repairs to the interior of the building and entrances to meet ADA design requirements. Forensic services involved: (1) reviewing the buyer's report claiming \$500,000 in remedial costs; (2) inspecting the facility; and, (3) analyzing the law to determine if the owner/seller needed to remove architectural barriers. Services also included assisting the owner's/seller's legal counsel in negotiations with the buyer's legal counsel to resolve differences and achieve an amicable settlement that would result in the successful sale of the building. Similar to the services for the county described above, these expert forensic

architectural services were preemptive so that parties would not have to enter legal action to resolve matters.

Case 3: Support of an intervenor in case to disallow a dangerous ramp to remain. The interested party, aware that a ramp (at a newly acquired lease space in building by the franchisee of a national restaurant chain) was constructed too steeply, engaged the expert forensic architect to assist with intervention if the state building officials decided in favor of allowing the steep ramp to remain. The site is in a small town's historic district. Therefore, any architectural modifications to the exterior of the building are required to be in accordance with historic guidelines as promulgated by the U.S. Department of the Interior. The site was inspected and the state-approved drawings indicated, erroneously, that the ramp was compliant with building codes. Calculations were made including a graphic demonstration of dangerous shift in center of gravity of a wheelchair user on the ramp, proving that the extreme steepness was non-compliant with both state building codes and federal ADA design standards. The ramp was not only non-compliant, but also extremely dangerous for people who use wheelchairs. After the state proceeded to approve the variance to state code, the forensic architect was then engaged as an expert in assisting the legal effort to appeal the state ruling. Also, the expert forensic architect assisted in filing a complaint with the U.S. Department of Justice. Unlike the two previously described cases above, this case has gone to court. The architectural expert continues to assist with this case both in the state court system as well as on the federal level through the U.S. Department of Justice.

Architectural, Construction, Design

C2 Richard III: Wounds and Weapons

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The goal of this presentation is to demonstrate how modern forensic techniques used for tool mark analysis can be applied in determining how injuries were sustained to a 500-year-old skeleton; and will combine techniques from forensic engineering science, forensic pathology, and archaeology to understand the wounds and weapons used to cause the injuries found on the King's skeleton. Additionally, discussion will involve the metallurgy of medieval armor and its ability to resist the penetration of arrows. A series of tests have been conducted on steels with differing carbon contents and heat treatments to understand the relationship between steel microstructure and the penetration ability of bodkin arrows that were used in medieval longbows.

This presentation will impact the forensic science

community and be especially relevant to forensic engineers, pathologists, anthropologists, and all others who have an interest in tool mark analysis and the relationship between tool marks and weapons.

Richard III was King of England between 1483 and 1485. He is perhaps best known today by the characterization presented by Shakespeare in Richard III — a stunted humpback with a scheming, vicious persona.

In 1485, King Richard III rode out from Leicester with his army to fight against Henry Tudor, latterly King Henry VI of England. The battle was fought at Bosworth Field around 20 miles from Leicester. By the standards of the time, Bosworth was a short battle lasting only around two hours. The battle reached its end when Richard III was killed.

Of all the monarchs of England since 839, Richard III was the only one whose final resting place was unknown. One legend had it that his body had been thrown into the river Soar and was lost forever. Other tales told of a church ruin with a plaque on a column claiming to mark his grave.

In 2012, the University of Leicester was given permission by Leicester City Council to excavate a site — a council car park — that, from scrutiny of ancient maps, could well be the site of the Greyfriars church where Richard may have been interred. It is not usual archaeological practice for a dig to be initiated to look for the remains of an individual.

Despite limited funding and over 500 years of lost records, the very first trench dug in the car park exposed a skeleton. Two further trenches revealed the outline of the ancient church and confirmed that the skeleton was buried in the choir of the church, a mark of high status. Moreover, the skeleton showed pronounced scoliosis, a curvature of the spine.

Carbon dating showed that the skeleton was of the correct age to be Richard III. Mitochondrial DNA testing on his descendants proved beyond reasonable doubt that the skeleton was indeed that of Richard III. The results of the find were announced by the University of Leicester to worldwide media coverage in February 2013. In the United States, this is often referred to as “the find of the King in the car park.”

The penetration of armor by arrows was an important part of the development of plate armor. The metallurgy and microstructure of the armors that were produced in the late medieval period were highly variable with a range of microstructures and differing hardnesses. A series of experiments have been conducted using plate steel with differing carbon contents and different heat treatments to better understand how armor penetration was related to the underlying microstructure. High-speed video has been used to image the penetration mechanisms. This presentation will discuss how weapons and armor are related to conclusions about the way in which Richard III was killed.

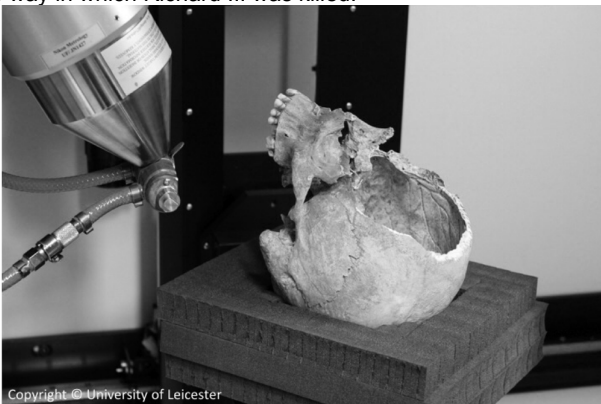


Figure 1: The skull of Richard III mounted in the micro-computed X-Ray tomography scanner.

C3 Front Occupant Injuries From Interior Intrusion by Rear Occupants

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The goal of this presentation is to present case studies that show some consequences of unrestrained adult-sized rear occupants that can cause intrusion into the frontal occupant compartment and cause fatal loading to the front seat occupants.

This presentation will impact the forensic science community by presenting case studies that demonstrate that not only heavy cargo, but also adult-sized unrestrained rear seat occupants, can have adverse consequences for the chance of survival of front seat occupants regardless of their seat belt use.

State law in Arizona requires front seat occupants to be seat belted. Children under 16 are required to use seat belts regardless of where they are seated, and children under five years must be seated in a child safety seat/booster. There are not any seat belt regulations on rear seat occupants who are 16 years or older.

A mid-sized Sport Utility Vehicle (SUV) traveled left of center and was involved in a head-on crash with a full-sized pickup as shown in Figure 1. From the crush, scene evidence, and the pickup Crash Data Recorder (CDR), the SUV's Delta V (change in velocity) was 36 to 38 mph and the pickup's Delta V was about 29 mph. Since the vehicles were both stopped at the point of impact, their speeds at impact approximately correspond with their Delta Vs.



Figure 1. The vehicles at rest.

The SUV's rear seat occupants were high school-aged, but their weights were in the 200 to 250-pound range. As seen in Figure 2, upon impact, the unrestrained rear seat occupants moved forward into the front seat backs. The seat backs were grossly deformed forward, intruding into the front occupant compartment to the degree that fatal injuries would be expected for the front seat occupants, regardless of seat belt use or airbag deployment.

In effect, the rear seat occupants used the seat backs and front seat occupants to dissipate their relative motion within the SUV. Therefore, had the front seat occupants been seat belted, the restraint loads for the front seat occupants would have more than doubled due to the additional unsecured mass of the rear seat occupants.

Interestingly, children were seated in the pickup's rear seat. As shown in Figure 3, even the mass of an eight-year-old is enough to cause sufficient seat back deformation to add to the restraint loading on a seat-belted front seat occupant.



Figure 2. Occupant compartment intrusion in SUV.



Figure 3. Seat back deformation in pickup.

Clearly, it would not only be advantageous to secure cargo, but it would also seem highly beneficial from a public safety or societal cost point of view to advocate that rear seat adult-sized occupants be secured. This not only for their own safety benefit, but it is also in consideration of the safety of the front seat occupants. Indeed, this case study exemplifies how the front seat passengers might sustain serious and/or fatal injuries, caused unnecessarily, by the rear occupant interior intrusion into the frontal compartment.

Rear Occupants, Intrusion, Seat Back

C4 Low-Energy Bone Fractures: Part II

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The goal of this presentation is to review the factors associated with reduced bone toughness and facilitate reconciliation of low-amplitude forces to seemingly disproportionate bone fractures.

This presentation will impact the forensic science community by showing forces calculated from reconstruction of low-velocity accidents or otherwise atraumatic incidents may appear incommensurate with the accompanying human injuries. This can lead to uncertainties regarding reconstruction mechanics or incident events. Bone fractures accompanying low-energy incidents are becoming more prevalent and are gaining greater attention. This presentation will review the factors linked with reduced bone fracture thresholds and thereby help the forensic investigator reconcile seeming incongruities between low-energy events, reconstruction derived forces, and accompanying bone fractures.

The accident reconstructionist/human injury analyst quantifies event kinetics, calculates associated impact forces, and links these forces to the resulting injury by using probabilistic methods. Although the magnitudes of the event-related forces are generally proportional to the degree of injury, the relationship between force and injury in low-energy events is often nonlinear. Such disparities can lead to uncertainties regarding the validity of the reconstruction, accuracy/inclusiveness of the events considered, or skill of the investigator.

These disparities may be partially resolved by considering the factors governing bones' fracture resistance. Bone is both a calcium-ion reservoir and a dynamic mechanical load-bearing organ. These vital functions are enabled by a time-varying composite biomaterial with biologically governed micro and macro structures. The mechanical competence of normal healthy bone is optimal; material or structural departures from normal will commonly result in compromised load-bearing capabilities and increased fracture susceptibility.

Bone mass, critical for fracture resistance, cannot be gained or maintained in the absence of essential building materials. Bone is a composite material made chiefly of protein (collagen) and mineral (calcium hydroxyapatite). Contemporary diets foretell reduced bone toughness because the "Pepsi® generation" eschews milk for carbonated beverages. Dietary calcium intake has thus been notably deficient.

Bone mass abnormalities are also linked to sex hormone inadequacies, most commonly evident in post-menopausal women. Menopause related estrogen loss precedes rapid bone loss due to high bone turnover characteristic of osteoporosis. Anti-osteoporosis drugs reduce turnover and bone loss, but new evidence suggests that their long-term use reduces fracture resistance due to oversuppressed turnover and unrepaired bone microdamage. This is important for America's aging baby boomer population and the four million adults currently taking anti-osteoporosis drugs.

Lifestyle changes also predispose bone to low-energy fracture. Bone mass increases rapidly during adolescence, peaks late in life's second to mid-third decade, then slowly declines. Exercise-induced bone mass increases during adolescence may persist (to a reduced degree) throughout life; however, inadequate skeletal loading during adolescence prevents attainment of peak bone mass. Persistent inactivity in adulthood results in "withdrawing bone" from a smaller "bone mass nest egg" and critical fracture thresholds are therefore achieved earlier. Reducing sunlight exposure also reduces blood levels of the active form of Vitamin D, critical for calcium absorption in the gastrointestinal tract, thereby further exacerbating calcium deficiencies. Current studies show mild Vitamin D deficiencies even in otherwise well-nourished adults. Extreme cases of Vitamin-D deficiency are associated with loss of bone stiffness and markedly increased fragility.

Clinical reports note the growing incidence of low-energy bone fractures in younger middle-aged pre-menopausal women with otherwise normal diets, sufficient exercise, and normal bone mass. The etiology of these fractures is unclear, but recent findings suggest abnormal collagen crosslinking that may be heritable. This is currently a field of active investigation.

To summarize, reconciliation of low-amplitude forces accompanying seemingly atraumatic incidents with resulting bone fractures requires detailed consideration of the material and structural aspects of the involved bone. This begins with a patient history, including prior diet, history, surgeries, lifestyle, and medications. Noninvasive diagnostic tests assessing bone quantity are essential; needed information may exist in current medical records. Missing information may be obtained from various assessments performed on bone samples, i.e., bone biopsies to quantify: bone structure, material composition, and microdamage. The forensic investigator should consider these and other factors to help understand the relationship between low-energy events and accompanying, but seemingly disparate, bone fractures.

Low-Energy, Bone Fracture, Bone Quality

C5 The Misuse of Daily Activities in Understanding Occupant Response in Motor Vehicle Collisions

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The goal of this presentation is to educate attendees regarding motor vehicle injury biomechanics.

This presentation will impact the forensic science community by providing understanding of injury parameters in motor vehicle collisions.

Over the last 20 years, there have been numerous papers published that report acceleration values on occupants and Anthropometric Test Devices (ATDs) in full-scale vehicle impacts, sled tests, amusement park rides, activities of daily living, and other events. These experiments often involve measurements at just a few locations, or even a single point, on the test subject or device. It has become common for investigators to take the values from these experiments and compare the forces on the vehicle's occupants to the accelerations on the vehicle or to the forces in other biomechanically dissimilar events, such as daily activities.

The U.S. National Highway Traffic Safety Administration (NHTSA) conducts numerous full-scale staged crash tests on a variety of vehicles and ATDs. While the NHTSA tests are typically conducted at higher speeds than observed in volunteer tests and the resultant kinetics are different, the kinematics can be compared biomechanically.

The data from these tests reveals that the measured maximum accelerations, loads, and moments for the head, upper neck, lower neck, chest, pelvis, left femur, and right femur are not the same. Additionally, the values vary in time and involve both minimums and maximums. Figure 1 is adapted from an NHTSA rear impact test on a 2000 BMW 328. The data is similar for hundreds of other rear impact tests. As the data demonstrates, the peak values for the upper neck, the lower neck, and the chest occur at different times. The data demonstrates that even if the acceleration values on the vehicle are known, the acceleration values on the occupant cannot be determined with accuracy and that one value on the vehicle does not describe the event in terms of the occupant.

Even if the change in velocity of a vehicle is known, Freeman has analyzed peak acceleration values of human subjects' heads as reported in 24 different studies.¹ Freeman showed that measured peak head acceleration values can vary by 600 percent for the same change in velocity under controlled experimentation.

The current literature has been reviewed and demonstrates that the use of a single acceleration value, or a dissimilar set of

forces, does not provide the same resultant acting on a vehicle's occupant during a collision. Furthermore, this approach is not biomechanically relevant to understanding injury potential in a motor vehicle collision. The kinematic response of an occupant in a motor vehicle collision typically does not replicate the motion in a daily activity such as sitting in a chair or jumping off a stool.

In 1994, McConnell et al. demonstrated the fallacy of using a single value in their 1993 paper on volunteer rear-impact testing.² Their paper showed a constellation of resultant accelerations at the top of the neck. This demonstrated that a rear impact was not similar to a daily activity. McConnell described the motion in a rear impact as a "unique kinematic pattern."

In 1994, Szabo et al. conducted volunteer impact tests involving rear impacts.³ Szabo demonstrated that the motions of the head, shoulder, wrist, and knee were all different. This study established that the angular displacement between the head and torso experienced by the volunteers was not repeatable and showed significant variation. This study showed that the peak lumbar accelerations were different than the peak cervical accelerations which were different than the head resultant accelerations. Additionally, the maximum and minimum values were not repeatable.

In 1998, Kroonenberg, et al., conducted 43 sled tests using nineteen subjects.⁴ Significant variation was demonstrated in head center of gravity displacement, T1 displacement, head angle relative to T1, head lag, resultant head acceleration, head angular acceleration, T1 resultant accelerations, upper neck shear forces, upper neck axial forces, upper neck torque, and head restraint impact forces.

Numerous other studies have demonstrated both the lack of repeatability of acceleration values among subjects and variation in values applied at different locations on the body. The data and relevant physics demonstrate that the accelerations involved in activities of daily living cannot be compared to the accelerations involved in a vehicle crash.

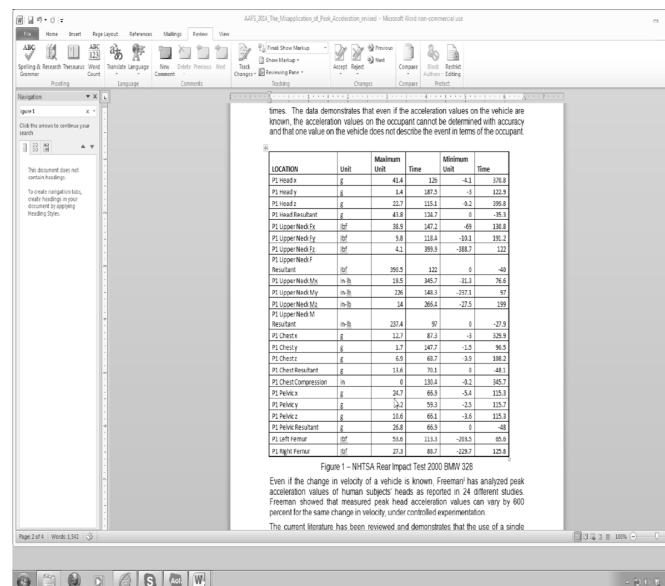


Figure 1. – NHTSA Rear Impact Test 2000 BMW 328
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Injury, Acceleration, Biomechanics

C6 Forensic Engineering Investigation of a Skiing Loss of Control Fatality

A.K. Aleksander, PhD, Aleksander & Associates, PA, PO Box 140558, Boise, ID 83714*

The goal of this presentation is to assist the attendee in understanding the investigative process and in becoming familiar with the terminology and critical factors that affect such an investigation.

This presentation will impact the forensic science community by increasing awareness of the fact that skiing fatalities, though relatively rare, are difficult to investigate, due to the inherent variability of the pertinent physical parameters of the winter terrain, as well as human cognitive behavior, dynamic performance, and the particular circumstances.

A 15-year-old girl befriended a 13-year-old girl at gymnastic classes and invited her for a weekend at the family's mountain resort residence. The older girl was a seasoned racer who had been coached for some years and competed in many races throughout the mountain west, particularly at ski areas in California.

She owned several pairs of skis, which were of a recent design.

The younger girl had had few opportunities to ski, but indeed owned an older pair of skis. The parents of the younger girl were hesitant to let her go, but after assurances from the parents of the 15-year-old that they would be with the girls at all times, agreed.

On a Saturday morning, the girls were dropped off at a major western ski resort while the parents went off to a real estate seminar. The girls got on the first chairlift at the base of the mountain, and proceeded to ski down an intermediate slope (marked as a family ski zone, and marked with slow speed banners), a distance of approximately 1,400 feet. At that point, the 13-year-old lost control, veered across the hill, and hit an island of trees. She was unresponsive with severe injuries, and although evacuated by the ski patrol and transferred by helicopter to a local hospital, she died of the injuries that evening.

The investigation focused on why this accident occurred, and identified the contributing factors.

Skiing as a sport has evolved through several significant generations of technical innovations. These include changes to basic concepts in ski design, most notably the length and side-cut profiles of the skis. These side-cut changes affect the turning characteristics of the ski. The skis the decedent was using were not her own, but lent to her by the elder girl. This fact, plus terrain and common skiing errors, will be examined in more detail. These differences between the older traditional "straight" skis, and the more recent "shaped," or "parabolic," or similar side-cut skis will be discussed, as they likely affected the outcome of this case.

Witness statements, photos, photogrammetric techniques, and site investigations helped in determining the sequence of events leading to the fatal crash. Also, ski testing and an evaluation of the ski design parameters further clarified the probability of a condition sometimes referred to as "railed edge" or "back seat," terms that describe the shifting of the center of gravity onto the rear half of the

ski length. This condition tends to freeze a person in an awkward position, resulting in unintended acceleration and loss of directional control. The performance characteristics of specific skis must be learned and are not necessarily readily transferable to other types of skis. This is one of the key findings in this case.

The case was litigated, and the results will be discussed. Ultimately, the very nature of gravity sports predisposes one to an elevated level of hazards that must be mitigated by training, caution, and adherence to basic safety guidelines. Parental oversight is one of those cautions.

Skiing, Skis, Ski Patrol

C7 Multiple Vector Impact Crashworthiness Affecting Restrained Vehicle Occupants: Ejection Caused by Predictable Seat Belt and Seat Failure

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After attending this presentation, attendees will learn the science and engineering involved in investigating at-scene and vehicle evidence in determining the foregoing failures in a foreseeable multi-impact collision, as well as how static and dynamic testing of the vehicle also demonstrated the predictability of the ensuing severe injuries in what should have been a readily survivable collision.

This presentation will impact the forensic science community by demonstrating the significant safety hazards affecting millions of motorists under similar impact conditions. This study especially highlights the emerging safety issues of second- and third-row seating in utility vehicles, as well as the unreliability of various occupant protection systems.

Vehicle frontal and side-impact protection is required by federal law. Technological advancements of seat belts, airbags, and other forms of frontal and side-impact protection have been achieved in response to these requirements. Despite decades of proposed rulemaking and published research on how and why there is a need to provide vehicle crashworthiness in dynamic rear and rollover collisions, there are no effective U.S. government or auto industry occupant protection requirements for rear impact or rollover. Consequently, there has been far less advancement made in the crashworthiness of most production vehicle seats, seat belts, and structures to insure occupant-protection in such collisions. It is common for foreseeable multi-impact collisions to occur, such as frontal impact followed by rear impact followed by rollover. Vehicle occupant restraints, such as seat belts and seats, must function properly to reduce interior contact and prevent ejection, not only during frontal and side impact, but also during rear impact and rollover; however, the latter two are not dynamically evaluated during normal vehicle development or safety testing. During rear and rollover impacts, various predictable seat belt and seat safety defects can cause vehicle occupants to be extremely vulnerable to ejection and injury.

This is a case study involving accident reconstruction, biomechanical analysis, vehicle design, and crashworthiness evaluation, as well as static and dynamic testing, to prove how and why ejection and injury of a belted occupant occurred in a minor to moderate multi-vector collision, and how it would have been prevented utilizing existing technology. A utility vehicle containing three restrained occupants incurred a moderate frontal impact with airbag deployment, followed by a low to moderate rear impact,

followed by a partial rollover onto one side of the vehicle. There was no loss of occupant survival space as a result of any impact. The driver and left rear occupant escaped without significant injury, because they were protected by their restraints in the frontal impact, and their seats generally remained upright, sufficient to prevent ejection during the rear impact and rollover. The initially restrained center rear occupant loaded the belt, had no detectable crash injuries, and made no detectable injurious contact with the vehicle interior during the frontal impact, but was ejected from the belt and seat and was severely injured due to contact with the road surface as a result of the rear impact and rollover. There were predictable failures of the rear seat and seat belt during the rear impact and rollover which affected occupant retention and protection. There was multiple evidence of seat belt usage, as well as catastrophic failure of the seat belt and seat during the rear impact. Static and dynamic tests were utilized to validate vehicle and occupant dynamics and to evaluate rear seats, head restraints, and belts under controlled scientific conditions. Production seats and belts were compared with alternative design seats, with and without belts. Seats repeatedly showed catastrophic failure that precisely matched the forensic evidence in the subject vehicle. Belt buckle failure and slackening have been demonstrated in dynamic testing and real-world collisions, and have been published in the technical literature since the 1960s.

Rear Impact and Rollover, Seat and Belt Failure, Occupant Ejection

C8 The Failure of a Bungee Drop in an Amusement Park

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After attending this presentation, attendees will understand the failure mechanism of a freely dropping ride called the "bungee drop" in an amusement park.

This presentation will impact the forensic science community by introducing the fatigue failure of the rolling bearings installed at the failed bungee drop that caused the death of one rescue worker and also by introducing various types of operation modes that can cause the failure of a bungee drop.

A moving frame installed with 24 seats for the ride traveled upward along the main structure using compressed air through a cylinder and dropped freely at the top of the bungee drop, at a height of 42.3m. Sixteen urethane wheels were installed onto the moving frame to guide it along the main structure. The weight of the moving frame itself was balanced using a counterweight through sheaves, and the compressed air needed to sustain and overcome the weight of the passengers was modulated by pneumatic shut-off valves operated by solenoid valves. The compressed air was supplied by two screw-type compressors of 110kW each with one set unloading pressure at 8kg/cm and the other at 9.5 kg/cm².

There were two accidents at the bungee drop. In the first accident, the moving frame with 15 passengers stopped abruptly in the middle of the main structure at a height of 27m. When the rescue team was trying to rescue the passengers from the moving frame, the frame dropped abruptly, causing one rescue worker to fall to his death. All of the urethane wheels with bearings for the guide of the moving frame were investigated. The urethane wheels were severely worn out, and some were octagonal as a result. In the roller bearings of the urethane wheels, various types of damage, such as fatigue, wear, and corrosion, were commonly seen.¹ With

fatigue damage, flaking that initiated from micro cracks below the surface was noted at the roller and inner ring. There was contact-trace evidence at the surface of the rod that was installed onto the piston in the cylinder. Conclusions from an investigation were that the moving frame was in an unstable equilibrium state that would be broken with a small disturbance load when the rescue teams were in operation.² The unstable equilibrium state was maintained by the weight of the passengers, the frictional forces caused by the urethane wheels, and poor lubrication of the rod.

A second accident happened nine years after the first accident. The moving frame with 12 passengers dropped abruptly as it was moving upward. It stopped at a height of 53cm due to the operation of the emergency brake system. Some passengers suffered spinal fractures. All the compressed air system equipment was investigated by varying the compressed air pressure. The moving frame was operated by varying the compressed air pressure as well, and all of the test cases were recorded by a video camera to analyze the time difference of the moving frame at each operation step. Conclusions of this investigation were that the moving frame dropped because only one air compressor, set to an unloading pressure of 8kg/cm, was operating while the other air compressor, set to an unloading pressure of 9.5kg/cm, was not operating at the time of the incident. After the investigation, it was recommended that a control system for the bungee drop should be installed to prevent the start-up of the moving frame, given the poor state of the compressed air system.

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Bungee Drop, Unstable Equilibrium, Flaking

C9 A Study on the Possibility of the Driver's Injury When Two Cars Collide With Their Side Mirrors

Sungji Park, 139 Jiyang-ro, Yangchun-gu, Seoul, SOUTH KOREA*

After attending this presentation, attendees will understand how to determine drivers' injuries in a specific type of traffic accident.

This presentation will impact the forensic science community by providing information on determining the probability of driver injury as a result of side mirror impact between two vehicles.

Recently there have been many people who have received monetary compensation for claiming injury after being the driver in a traffic accident that involved only slight contact between the side mirrors of the two cars. This type of accident, called a "moral hazard," has spread rapidly across the whole country of South Korea. Therefore, the possibility of injury to the driver from these type of accidents is examined according to logical bases.

Testing was conducted to analyze the possibility of injury to the driver in a case where the two cars slightly contacted with their side mirrors. In the testing, a pendulum was prepared to impact the side mirror of a car. The prepared pendulum was 25kg, 3.8 meters in length, and rotated at the upper end around a pin joint. An acrylic plate which was 20mm in thickness was attached to the pendulum hammer. The pendulum was rotated upward 60 degrees and then released so that the hammer contacted the side mirror of the car. This test was repeated ten times for the frontal direction and the rear direction. During the testing, the glass of

the mirror separated from the side mirror housing and also the mirror cover was fractured several times. So, the impact was more severe than the actual collision of the two side mirrors of two cars, where the damage to the mirrors was less.

A triaxial acceleration sensor was applied for recording vibration according to ISO2631. In the case of the frontal impact, the vibration transmitted to driver was measured at 0.4 Maximum Transient Vibration Value (MTVV) and 0.7 Vibration Dose Value (VDV), and in case of the rear impact, the vibration transmitted to the driver was 0.15 MTVV and 0.23 VDV.

For comparison, testing was also conducted by measuring the vibration level when the driver's door on the car was closed. The door was opened 40cm and closed at a velocity of about 0.35m/s (not slamming the door). The vibration levels measured in the door closing test were 0.5 MTVV and 1.0 VDV.

The test results indicate that the vibration level transmitted to the driver when side mirrors collide with each other was lower than when the driver's door is closed. It was concluded that it would be difficult for the driver to receive any injury when both side mirrors of two cars collide.

Injury, MTVV, VDV

C10 Electronic Crash and Injury Causation Analyses

Jacqueline Paver, 501 Meigs Road, Santa Barbara, CA 93109; and Donald Friedman, BS, 501 Meigs Road, Santa Barbara, CA 93109*

After attending this presentation, attendees will understand how to determine and apportion motor vehicle accident and/or injury causation using system analysis.

This presentation will impact the forensic science community by demonstrating the use of previously unavailable information to analyze accident and/or injury causation.

Hypotheses: Any crash is related to a combination of roadway factors, vehicle defects, and/or driver errors. The ever-increasing electronic control of vehicles has its benefits and limitations. Modern vehicle Drive-By-Wire (DBW) systems can have life-saving outcomes because they can identify and take action (e.g., deploy airbag) based on potentially dangerous roadway factors, as well as vehicle and/or driver errors. Most of the time, the DBW systems save lives; however, some DBW commands can be fatal (e.g., misinterpreted data causing a crash or preventing airbag deployment causing injury). A system analysis that utilizes the stored vehicle control module data, in conjunction with physical evidence and Crash Data Retrieval (CDR) download, enables the forensic engineer to quantitatively apportion causation and provide proof of roadway factors, vehicle defect, and/or driver error.

Objectives: A system analysis methodology to: identify, collect, and analyze the data stored by vehicle control modules, and interpret control module events, faults, limits exceeded, and Diagnostic Trouble Codes (DTCs) to determine and apportion accident and/or injury causation.

Background: Modern vehicles are operated by increasingly sophisticated DBW systems. Sensors measure vehicle and occupant data. Data acquisition systems collect the data. Control modules analyze and store the data and trigger or inhibit vehicle-controlled corrections and/or safety system deployment. Communication networks transmit the data and commands. Currently, the system records up to an 18-second time history of vehicle data (e.g., accelerator pedal, throttle and brake position, steering angle; pre-deployment and deployment accelerations; frontal, side and window curtain airbag deployment and timing parameters) and occupant data (e.g., belt usage, seat

position, and weight). The vehicle control modules analyze the data, identify the events, faults, limits exceeded, and DTCs, then take action (e.g., command airbag deployment or nondeployment).

Systems Analysis Methodology: (1) Obtain and review Product Definition Documents (PDD) and DTC interpretations; (2) Scan the sensing and diagnostic module (SDM) for DTC's with the Tech 2 tool; (3) Scan the control modules for stored data with the NEO VI tool; (4) Analyze stored data to identify events, faults, limits exceeded and DTC's; (5) Reconstruct the accident from the physical evidence and electronic data; and, (6) Render opinions about accident and/or injury causation.

Downloadable Data: The content and coding of the downloadable CDR data is well documented. Presently, however, only selected data of these 40 or more vehicle control modules can be downloaded. The downloadable data is by no means all of the data stored in vehicle control modules. The CDR downloads include only the data that the manufacturers want the consumer to have.

Confidential and Proprietary Data: For years, sensors, data acquisition algorithms, data processing algorithms, control module events, faults, limits exceeded, and DTCs in the SDM were classified as confidential, proprietary, and inaccessible with commercially-available tools. Only recently, the courts ruled that the data and its interpretation are the property of the vehicle owner.

Example of a System Analysis of the Physical and Electronic Evidence in a Real-World Crash: The available CDR data in a 35-mph barrier crash documented driver belt usage and airbag inflation; however, belted passenger airbag deployment was prevented due to the "detected" presence of a small belted adult passenger. Physical evidence of vehicle contact with a redirecting Jersey barrier moments before frontal barrier impact suggested a vertical lift of the 170lb adult passenger from his seat. Access to *confidential data* revealed the time history of instantaneous passenger weight. Analysis confirmed that the passenger weight was an instantaneous function of that vertical lift. The airbag deployment algorithm was confirmed as the defect. The alternate design utilized an averaged occupant weight algorithm.

Conclusion: A systems analysis methodology is presented that utilizes the stored vehicle control module data, in conjunction with physical evidence and CDR download to enable the forensic engineer to quantitatively apportion causation and provide proof of roadway factors, vehicle defect, and/or driver error.

Algorithm, Microprocessor, Diagnostics

C11 Experimental Study of Seat-Back Recliner Sudden Failure and Effect on Rear Child Injury in Rear Impacts

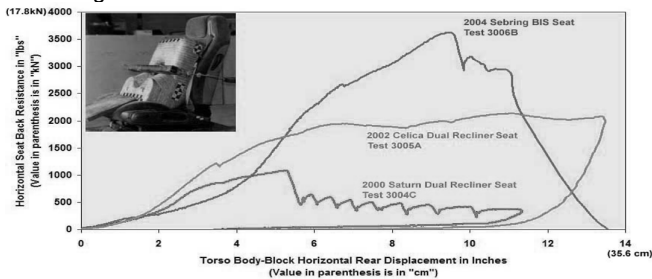
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After attending this presentation, attendees will learn about an efficient "quasi-static" test method for measuring vehicle front seat ultimate rearward force resistance, deformation levels, and "reliability" of seat support regarding a rear child in rear impacts.

This presentation will impact the forensic science community by demonstrating a method that provides a simplified means for evaluating seat safety reliability in rear impacts.

Similar to the shoulder belt in a frontal impact, the seat back is the primary restraint device available to protect both the front

adult and children seated behind in rear impacts. "Rear impact" crash test multi-variable studies, discussed at previous American Academy of Forensic Sciences (AAFS) meetings and published in engineering society proceedings have demonstrated that, among other factors, the stronger front seat systems, such as the dual recliner "non-belt-integrated" types and the "Belt-Integrated-Seat" (BIS) types, generally provide less penetration into the occupant space of children seated behind the weaker "single recliner" yielding seat, and as such result in a safer environment for both the front adult and the rear child.¹⁻³ Experimental results have been shown to be consistent with statistical accident database studies conducted independently by others, such as Jermakian et al. of the Childrens Hospital of Philadelphia, and Mango and Garthe.^{4,5} However, crash tests and "Quasi-Static" (QS) ultimate seat strength and deformation tests have shown that, in some instances, the rear child safety value of a potentially stronger and less rearward yielding seat can be voided if seat components like the recliner seat back adjustment mechanisms suddenly fail or give way, such as when "recliner locking gear teeth" distort and slip over one another. These types of failures nullify the reliability and safety benefits of the stronger seat back restraint factors.



Unfortunately, there are no federal requirements to measure reliability of ultimate seat strength and deformation data, as has been done by the private sector since the early 1990s. One method uses a torso-body-block device, similar in weight to a 95th percentile male, to more realistically load the seat system QS and enable efficient duplication of sudden failure deficiencies not found by limited test approaches. This QS method data also enables estimates of torso "Peak G" dynamic loads and has been shown to correlate with actual crash test and "real world" accident case findings. The figure above shows a photo inset of the torso-body-block test setup and data from "ultimate seat strength and deformation" tests of three different seat systems; one seat is the much stronger 2004 BIS Sebring seat, and two are the less strong Dual Recliner (DR) types. The result for the 2000 Saturn DR-type seat illustrates a sudden load failure "drop-off" in seat load when recliner gear-teeth slipping occurs, making this system an unreliable safety device. More important, since the area under the load-deformation curve is proportional to energy absorption, the seat with the sudden load failure ultimately absorbs much less energy than the other two seats and, as a result, put both the front adult and rear child at risk of enhanced injury. In this study, the torso-body-block seat strength method is described and results of adjuster mechanism failures are shown and correlated with sled-body-buck crash tests to demonstrate consistency with actual cases involving rear-seated child injuries.

References:

1. Saczalski, K., Pozzi, M., Burton, J., "Strong vs Weak Seats; Analysis of "Matched Pair" Rear Impact Tests for Head & Neck Injury Risk Evaluations with Normal Out-of-Position Adults", Presented at the 60th American Academy of Forensic Sciences Meeting, Washington, D.C., February 18 – 24, 2008.
2. Saczalski, K., Saczalski, T., Pozzi, M., "Method for Predicting Rear-Impact Force Levels Associated with

Bumper Override and Sheet Metal Crush", Presented at the 64th American Academy of Forensic Sciences Meeting, Atlanta, GA, February 24, 2012

3. Saczalski, K., Pozzi, M., Burton, J., "Rear Seated Child Injury Risk Experimental Measures Related to Vehicle Front Seat Performance in Rear Impacts", Paper IMECE2009-10390, ASME, 2009.
4. Jermakian, J., Arbogast, K., Durbin, D., Kallin, M., "Injury Risk for Children in Rear Impacts: Role of the Front Seat Occupant", presented at the 57th AAAM Annual Conference, October 2008.
5. Garthe, E., Mango, N., "Standard & Integrated Restraint First Row Seat Performance in Rear Impact Crashes", NHTSA VSR/ESV Conf. Paper No. 11-0381, Washington, DC, June, 2011.

Rear Impact, Seat Strength, Quasi-Static Testing

C12 Forensic Engineering Investigation of a Steam Iron Electrocution

A.K. Aleksander, PhD*, Aleksander & Associates, PA, PO Box 140558, Boise, ID 83714

After attending this presentation, attendees will better understand the forensic engineering investigative process, the use of X-rays to determine defects, and the use of photography to document arc phenomena.

This presentation will impact the forensic science community by increasing knowledge of the necessary conditions for inducing ventricular fibrillation from a steam iron, which are that the person must contact a surface with a sufficient voltage potential, the person allows a path to ground, the iron must have a defect (or defects) that bypasses the internal pathways for current flow, and that there is a failure of the usual grounding provision for appliances that plug into a conventional outlet.

In a San Diego neighborhood in the early morning, a wife was ironing clothes using a conventional steam iron. A few minutes later, she suffered ventricular fibrillations, and died at the scene.

Initially, it was not clear that the iron had anything to do with the incident. However, the ensuing investigation showed that the iron was indeed defective and was the proximate cause of the death.

In this unfortunate case, all the necessary conditions were present to electrocute the user of the steam iron.

A systematic investigation found that there was a fault in the wire, a fault in the seal that separates the water chamber from the internal parts of the iron, and that a path to ground, except through the user, was not available.

The two-prong plug (rather than the three-prong usually found on irons) offered no separate ground connection from the appliance to the ground buss in the apartment wiring. Furthermore, an X-ray showed that the wires next to the plug were frayed and were in fact separated on the neutral (or return) side of the plug.

This meant the full 120 volt potential was conducted to the iron, with only intermittent, if any, current flow to the iron.

Internally, the seal that contained the water in the water chamber leaked. When the iron was in a vertical rest position, this leakage dripped water onto the connection from the energized circuit to the heating element embedded in the ironing surface.

It was further developed that the user of the iron was in the habit of wetting her finger and tapping on the heated surface of the iron to test if the iron was ready to use. In this case, even if the iron was cold to the touch, it was nevertheless a lethal hazard.

Given these conditions, a perfect storm existed. As the woman touched the iron, she inadvertently created a path

to ground, killing her. The presentation will show the details of the findings, including the X-rays showing the frayed wiring, the ineffective seal, and most interestingly, photos of the instantaneous arc created across the heating element to circuit interface. And, had there been a three prong-wire, this incident would not have occurred. A Fault Tree representation helps to evaluate the risk potential of similar devices.

Electrocution, Steam Iron, Fault Tree

C13 Forces in Vehicle Side-View Mirror Collisions

Russell L. Anderson, MS, PO Box 7185, Tempe, AZ 85281*

The goal of this presentation is to present force data associated with folding a vehicle's side-view mirror forward or backward.

This presentation will impact the forensic science community by presenting case studies that demonstrate the use of the force data to analyze actual or claimed interactions between the side-view mirror with another vehicle or a pedestrian.

This presentation includes the results of measured forces necessary to fold side-view mirrors forward and rearward and how they were used in two case studies involving vehicle and possible pedestrian interactions with vehicles' side-view mirrors. After attending this presentation, the attendees will understand the forces and accelerations involved in interactions with a vehicle's side-view mirror.

Case 1: This case arose from a sideswipe type accident in which contact was limited to only mirror-to-mirror contact between two vehicles. The side mirror of the subject vehicle was struck and folded forward, striking the A-pillar, as shown in Figure 1.

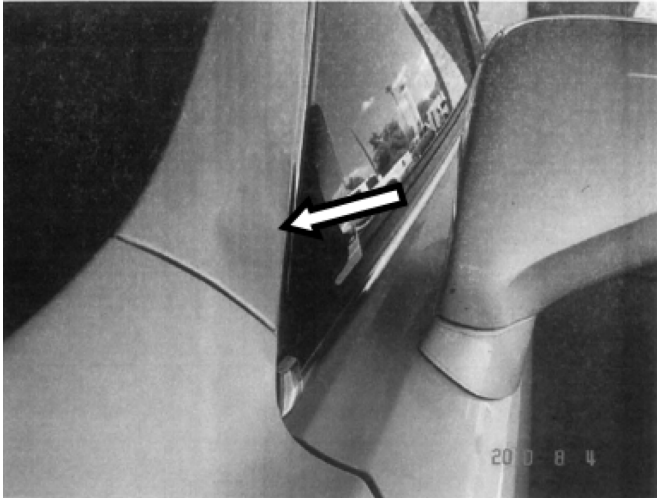


Figure 1: Damage due to mirror-to-mirror contact.

In order to determine the accelerations experienced by the vehicle and its occupant, three vehicles were used to measure the force required to fold their side-view mirrors forward. The force ranged between 9 and 12 pounds, which corresponded to about 1/50th of 1g or a Delta V less than 1/20th of one mph for the subject vehicle. For comparison, as shown in Figure 2, the longitudinal vehicle accelerations associated with normal driving were measured as a vehicle was driving on a normal roadway prior to a steering-induced rollover and were about 1/4th of one g. As such, the forces and accelerations which the vehicle and the occupant were subjected to were far less than those associated with vehicle vibrations while driving normally.

Thus, with contact limited to only mirror-to-mirror contact,

the occupant would not be able to discern the contact from the vehicle vibrations associated with normal driving, other than the sound of the mirror contact.

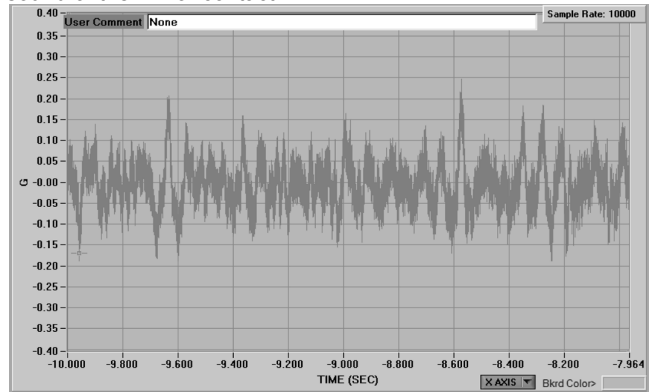


Figure 2: Longitudinal acceleration trace under normal driving conditions.

Case 2: This case arose from a vehicle versus pedestrian accident in which the pedestrian claimed that the vehicle's side-view mirror struck the pedestrian's right elbow resulting in injury. Video surveillance determined the speed of the vehicle to be between 11 and 13 mph as it passed the pedestrian. Interestingly, as shown in Figure 3, the height of the pedestrian combined with the pedestrian's forearm being raised holding a video camera was found to prevent direct mirror to elbow contact. In addition, surveillance video analysis showed that the mirror was not rotated rearward as it passed the pedestrian. The force necessary to fold the subject vehicle's mirrors was measured to be between about 11½ and 15 pounds. Certainly, had the vehicle's side-view mirror had significant contact to the bony structures of the pedestrian's arm as it passed at 11 to 13 mph, the side-view mirror would have been expected to fold rearward.



Figure 3: Surrogate demonstrating arm-to-mirror clearance

An examination of two case studies was performed. In these case studies, the amount of force necessary to fold the side-view mirror forward was measured to be between 9 and 12 pounds, which in Case Study 1 corresponded to vehicle accelerations that were far less than those which would be normally experienced while driving on a roadway and not discernible from the vehicle's normal vibrations. In Case Study 2, the amount of force necessary to fold the side-view mirror rearward was measured to be between about 11½ and 15 pounds, which, in addition to the height of the pedestrian's elbow/arm preventing contact, showed that the side-view mirror, would have been expected to fold rearward had it actually had significant contact with the bony structures of the pedestrian's elbow/arm.

Side Mirror Contact Forces, Injury, Case Study

C14 Investigation of Ear Witness Testimony With Regard to Sounds Heard During a Shooting Incident

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The goal of this presentation is to demonstrate the benefits of the application of established scientific knowledge to aid in the understanding of a complex situation.

This presentation will impact the forensic science community by affecting the way that witness interpretations of noises heard during shooting incidents are investigated.

The paper focuses on an actual case study to demonstrate the relationship between different noises reaching witness' ears during shooting incidents. Witnesses often misinterpret what they hear and this can have an impact on their testimony, charges filed, and the outcome of litigation.

The subject incident arose from a single-shot event. An armed suspect was holed up in a house that was surrounded by law enforcement officers. Two groups of three officers were located approximately 100 yards from the rear of the house, and 100 yards apart. The suspect discharged one round from his firearm in the general direction of the officers. The officers subsequently filed statements making contradictory claims about what they had heard, and what the implications of those sounds were. Officers in both groups said they heard a projectile whistle over their heads. Others said they heard a crack as the bullet passed over them, or perhaps a crack of a door banging on a house to their rear, or a bullet impacting a wall behind them. A search of the area revealed no spent bullet, and no impact damage to the walls of residences to the rear of the officers. It was cold and windy at the time of the incident, and there was considerable activity in the area due to the presence of law enforcement and their vehicles.

The question was, just what did/could the officers hear at the time of the incident? The model and caliber of the firearm used were known and there were two possible types of ammunition that could have been used. Weather data for the time of the incident was available. Two analyses were performed and the results combined so that what the officers could possibly have heard could be ascertained. The first analysis was to determine the time of flight of the bullet to reach the officers' location and the location of the residences behind them. This was accomplished using trajectory modeling software. The second analysis was to determine the time that the gunshot noise took to reach the witnesses' locations and the time that any bullet impact noise from behind the witnesses took to reach their ears. This was accomplished using established physical relationships for the velocity of sound at specified temperatures.

The results of these two analyses were tabulated and used to determine what the officers may have heard. It was established

that the gunshot noise (loud) and the bullet would take the same time to reach the witnesses. The bullet was subsonic at the time it reached the witnesses and, consequently, there would be no sonic crack to hear. Any "woosh" or "whistle" sounds would be minimal, and far quieter than the noise from the gunshot. It was concluded that the witnesses could not have heard a bullet passing close to their heads because any such noise would have been drowned out by the gunshot noise originating from the discharging firearm. Whatever noises the witnesses heard would have been before or after the bullet had been in their proximity.

Bullet Trajectory Modeling, Speed of Sound, Gunshot Noise

C15 Officer Fatally Shot in the Line of Duty: Murder or Suicide?

Michelle R. Hoffman, MS, 3720 E La Salle Street, Phoenix, AZ 85040; Carley C. Ward, PhD, Biodynamics Engineering, Inc, 3720 E La Salle Street, Phoenix, AZ 85040; Parris Ward, JD, Biodynamics Engineering, Inc, 17383 W Sunset Boulevard, Ste 290, Pacific Palisades, CA 90272; and Matthew A. Ivory, BS, 3720 E LaSalle Street, Phoenix, AZ 85040*

After attending this presentation, attendees will understand how a biomechanical analysis of fatal gunshot injuries can contribute to conclusions regarding manner of death.

This presentation will impact the forensic science community by demonstrating a case where an analysis of fatal injuries from a biomechanical standpoint provides insight into whether or not a person was murdered or committed suicide.

Summary of Case Facts: An on-duty police officer was found with a fatal gunshot wound. The medical examiner ruled the manner of death as suicide. A biomechanical analysis was conducted to determine if this was suicide or murder.

The officer was found in a vacant lot, lying on his back with his legs straight, his arms bent at the elbows, and his hands resting on his torso above waist level. His shotgun was on his chest with the muzzle below his chin.

The shotgun was a manually operated Remington® Model 870™ pump-action firearm loaded with department-issued federal 12-gauge, .50-caliber solid slugs with sabot. The slug was never found. Likewise, a recognizable impact between the slug and the asphalt or adjacent soil was never located. The wad was found approximately 35 feet from the officer's head.

Recoil testing with the gun held in two hands, but not against the body, demonstrated recoil distances from five to nine inches. Recoil testing with the gun on the ground (not held by a human) demonstrated a much greater recoil distance of 48 inches, estimated from video.

Injuries: The officer sustained a fatal gunshot wound to the head with the entry wound under the chin. Additionally, there was a patterned abraded laceration (2.5x1cm) just posterior to the entrance perforation. The bullet path ran through the mouth floor, tongue, palate, nasopharynx, skull floor, frontal lobes, and parietal skull with external beveling. The exit wound was over the right superior frontoparietal region. A second exit wound, from the wad, was just above and medial to the right eye. The direction of the bullet path with respect to the standard anatomic position was upward and from left-to-right. Postmortem radiographs showed no projectile in the head.

Analysis: Based upon the location of the gunshot wound, as well as the high energy of the weapon used, the officer would have been incapacitated and lost bodily control virtually instantaneously upon the shot being fired.¹

If he were standing when the shot was fired, his body would have collapsed vertically and would not have come to rest

as he was found at the scene. In order to reach his as-found position on his back from standing, his body's center of gravity (cg) would most likely have to have been traveling rearward with some initial velocity, such as being shoved rearward or initially running backward. If he were moving, the gun would not come to rest on his chest. His as-found body position is consistent with being moved by someone after the shot was fired.

If the officer was initially lying on his back at the time the shot was fired, his arms would not be in the as-found position had he pulled the trigger. His instantaneous loss of body control would not allow any significant force to counteract the gun's recoil and the gun would not likely have been found on his chest. Thus, his position and the gun's position are inconsistent with suicide.

The abrasion/laceration located posterior to the entry wound on the underside of his chin requires an interaction with a sharp object or a forceful interaction with a blunt object. The end of the tubular magazine under the barrel is not sharp and the force required to lacerate his skin would not exist in the suicide scenario, because of recoil. This abrasion/laceration means the gun was forcefully pushed under his chin and the recoil was then resisted, or the gun was moved rapidly toward his chin/neck as it was fired. Only a person positioned to control the recoil or a person shoving the gun with speed into the officer's chin/neck as the gun was fired could cause the magazine to abrade/lacerate the tissue. Thus, this abrasion/laceration was not produced in any potential suicide scenario.

Conclusion: A biomechanical analysis of the fatal gunshot wounds to the on-duty police officer indicates that his death was not a suicide, but a murder.

Reference:

1. Spitz WU and Fisher RS, Physical Activity Following Fatal Injury by Gunfire, in *Medicolegal Investigation of Death*, p. 266, 1980, Thomas Books, Springfield Illinois.

Gunshot, Fatality, Biomechanical Analysis

C16 3D Reconstruction of Non-Lethal Shooting Incidents Using Computed Tomography (CT) Scan Data

Parris Ward, JD, Biodynamics Engineering, Inc, 17383 W Sunset Boulevard, Ste 290, Pacific Palisades, CA 90272*

After attending this presentation, attendees will understand how X-rays and Computed Tomography (CT) scans can be used to model non-lethal shooting events on the computer.

This presentation will impact the forensic science community by demonstrating techniques for enhancing the accuracy of computer models used to analyze shooting events through the use of radiological data such as CT scans.

In creating a 3D model or animation of a shooting event on the computer, CT scans or other radiological data can be helpful in accurately depicting bullet paths through the body, especially in non-lethal shootings where no autopsy is performed.

Shooting events are often analyzed on the computer using 3D modeling and animation software because of its ability to demonstrate the interaction between objects and projectiles in terms of time and space. A computer model of the scene is created and mannequins are placed in the scene to represent the individuals involved. Bullet paths can be depicted as lines through space, from their point of origin to the objects they strike. When a person is hit, the path of the bullet through the body can be depicted with lines placed through a mannequin much the same way a pathologist places rods through a body. The lines representing the bullet paths are typically placed according to descriptions and measurements noted in the coroner's report, as well as autopsy photos of the

wounds. Of course, if the victim isn't killed in the shooting, there will be no coroner's report or autopsy photos. Thus, in non-lethal shootings, one has to rely upon other information in defining the bullet's path through the body. In this case study, CT scan data was used to determine a bullet path.

In this shooting incident, a police officer shot a suspect inside the suspect's home. According to the officer, he was hit over the head by an unknown assailant and fell to the floor when he entered the home. After a brief lapse of consciousness, he remembered someone dragging him by his head toward the back of the house while someone else pulled at his belt. He said he was able to get to his hands and knees and fired one round at the person in front of him. The suspect who was shot had a different recollection of events. He said he had been asleep in another room and walked into the dining room rubbing his eyes. He saw the police officer, heard a shot, and felt an enormous amount of pain. The suspect was shot in the abdomen and subsequently recovered from his injury.

There were no photographs taken of the wound, and medical records merely described the wound as occurring in the "right upper quadrant of the abdomen about midway from the umbilicus to the costal margin." Fortunately, a CT scan was taken before surgery. Both the entry wound and the bullet itself were visible in the scans.

The angle of the bullet path through the body was reconstructed from the CT scans. Using such data has certain advantages over autopsy findings. Whenever a bullet path through a body is replicated on the computer, there are limitations to the precision of such placements. A computer model of a person, or mannequin, is an idealization of the human structure. Human body types and proportions vary from person to person, thus a computer model cannot exactly match a specific individual's anthropometry. It also cannot articulate exactly like a human. The data found in the coroner's report has limitations too. The pathologist's measurements are typically taken while the body is on a table using a linear tape or rule rather than with a 3D measuring device. Yet, despite these limitations, a computer model can be produced that generally shows the direction and location of bullet paths through the body. When CT scan data shows the entry wound, the path, and the location of the projectile in the body, there is more information available to work from. There is better accuracy in terms of defining the location of both the wound and the projectile and anthropometric data unique to the individual is available, such as the dimensions of the chest and abdomen, which allows for increased accuracy in defining the angle of the bullet path. In this case, the angle of the bullet path helped to determine whether the suspect was standing or bent over at the time he was shot.

Shootings, CT Scans, 3D Animation

C17 Was That Car Used as a Weapon? Combining Reconstruction Skills to Answer a Critical Question

Michael L. Shirley, BSME, PE, 2290 County Road 23, Waterloo, IN 46793-9415; and John Nixon, MBA*, ARC, PO Box 66, Bippus, IN 46713*

The goal of this presentation is to highlight the benefits that multidisciplinary teams bring to an investigation.

This presentation will impact the forensic science community by affecting the way incidents involving firearms and automobiles are investigated by highlighting the overlapping technologies and demonstrating how the two disciplines complement one another when the experts work as a team.

This presentation focuses on a real world practical

example of a life-and-death struggle that developed from a routine traffic stop. When a law enforcement officer pulled over a lone driver following a minor traffic infraction on a suburban roadway, he did not expect a major struggle. However, after providing false identification, the suspect became aggressive and combative and the officer called for backup. The suspect was extremely strong, and the two officers used their Tasers® in an unsuccessful attempt to subdue him. The struggle lasted for several minutes, and the suspect eventually managed to get behind the wheel of his manual transmission car and move off at full throttle. One of the officers was in the vicinity of the front driver-side of the vehicle and, fearing for his safety; both officers fired shots from their handguns. Some bullets entered the vehicle and the suspect was fatally wounded.

Attorneys, acting on behalf of the suspect's estate, filed a lawsuit against the two officers and their employer. The plaintiff's attorney hired a shooting reconstruction expert who produced a report that indicated that the police officer at the front of the car was two to four feet away from the vehicle and, consequently, was not in danger at the time the officers fired their guns and killed the suspect. The plaintiff's expert report made a number of assumptions with regard to the incident and, importantly, the lone expert had very limited technical knowledge of vehicle mechanics and dynamics.

The defense experts conducted an investigation to determine if the police officer at the front of the vehicle was in any danger at the time the officers opened fire. The investigation first established the trajectory of bullets that entered the vehicle through the front windshield. Next, the officer was instructed to adopt his shooting stance and was positioned so that his pistol was coincident with the trajectory line of the bullet going through the windshield. This established the position of the officer to be approximately 16" forward of the driver-side front wheel of the suspect's car.

The next step of the investigation was to determine if the officer was in any danger while standing in this position. Witness testimony stated that the suspect applied full left lock as he pulled away in his car at full throttle. The vehicle was examined and the steering geometry measured. Calculations were performed to determine the turn radius of the vehicle at full lock. Industry test data were used to calculate the acceleration of the vehicle as it sped away from a standing start. The turn radius, vehicle acceleration data, and the location of the police officer were used to perform calculations. The calculations demonstrated that it would have taken less than 0.5 seconds for the suspect's car to reach the officer. Additionally, a practical demonstration was performed with the officer and the vehicle — it demonstrated that the driver-side front wheel of the vehicle would have run over the police officer.

The combined use of measurements, calculations, and practical demonstration was sufficient to prove that the police officer was in danger at the time of the shooting, and that the shooting was justified. The combined expertise of two engineering disciplines proved invaluable in this investigation.

Bullet Trajectory, Vehicle Engineering, Incident Reconstruction

C18 Evaluating the Impact of Reroofing on Increasing the Risks of Ponding of Water

Daniel M. Honig, PE, Structures Consulting Engineer, PO Box 125, Swarthmore, PA 19081*

After attending this presentation, attendees will understand some factors that affected and contributed to a roof ponding event, as well as how the selected materials, installation procedure, and localized drainage conditions caused the collapse of a recreation facility roof.

This presentation will impact the forensic science

community by illustrating how modification work done on a building can negatively impact the structural integrity of that building, using a case study. This presentation also highlights the importance of building code compliance, which often prevents building collapses as building modifications are made over time.

Ponding, the unexpected pooling of water, often occurs with catastrophic events and is a predictable condition in areas of heavy rainfall. The weight of rapidly congregating water on a rooftop can cause an existing structural roof system to deflect and progressively collapse during intense rainstorms.

In this instance, reroofing work completed on a recreation facility negatively altered the roof drainage capability of the building. The continuing adequacy of the rooftop drainage system was not properly reevaluated as part of the reroofing project, thereby leading to a significant structural roof failure incident approximately a year later during a rainstorm microburst. In this case, a localized portion of the structural steel support system of the roof collapsed due to ponding from a blockage created in the drainage system.

Prior to this ponding incident, the original roof drainage system had functioned properly for over twenty years since the time it was constructed.

The major factor contributing to the ponding failure was the installation of two-inch diameter drain inlets without proper debris-protection baskets during the reroofing process. The thickness of the drain inlets and the installation of an Ethylene Propylene Diene Monomer (EPDM) membrane within the inlets reduced the effective drainage diameter to approximately the size of a common bathtub or shower drain. This reduction in diameter significantly and proportionally reduced the effective drainage area of the roof drainage system by as much as 80%. In addition, no secondary or overflow roof drainage system was provided as per code. The new roofing material that was installed, including flute fillers and insulation, also created additional dead loads to be supported by the existing structural roof framing members.

While the roofing design of the original building would not have required emergency scuppers, the drainage system was altered, thereby making it inadequate to withstand even regular rainfall events. The significant drainage capacity reduction, in combination with the lack of protection baskets and the potential for blockage of the inlets, allowed the significant roof ponding and additional roof loading to occur. In this case, the reroofing contractor did not conduct a proper engineering review of the rooftop surface conditions as part of the completion of the reroofing job. While plumbing codes prohibit the reduction of drainage diameters on existing buildings, appropriate attention by installers to the impact of modification work on the structural integrity of a building could also help prevent such failures in the future.

Ponding, Roof Collapse, Reroofing

C19 Adaptations in Foundation Design: Have Improvements Occurred Through Science or Litigation?

Michael D. McDowell, MS, 24665 E Ontario Place, Aurora, CO 80016*

After attending the presentation, attendees will learn several important principles of geotechnical engineering and foundation design, including the history of foundation design changes in areas of expansive soil. Additional discussion will focus on common causes of foundation movement and historical data related to design changes and deep foundations. Special emphasis will be given to the Colorado experience and the potential effect of construction defect cases on current foundation design.

This presentation will impact the forensic science

community by presenting a history of the progressive nature of foundation design. The presentation will document advances in engineering design methodology and trends in foundation design over time. This presentation will also summarize data and testimony that suggest that significant changes in foundation design may correlate with an increase in construction defect litigation.

Expansive soils cause more damage to residential structures (and pavements) than any other natural hazard. Available data suggests that the effects of expansive soil are more costly than damage caused by earthquakes, floods, tornadoes, and hurricanes combined. Expansive soils can damage foundations by uplift as they swell with increasing moisture. The American Society of Civil Engineers estimates that a quarter of all homes may suffer damage caused by these soils. Because of these problems, this presentation focuses on the importance of providing adequate engineering designs that can reduce the risk of damage from expansive soils.

Swelling soils can lift and crack lightly-loaded footings and cause distress to floor slabs and interior finishes. To combat the negative effects of expansive soil, engineers began to modify foundation designs to reduce the risk of differential movement. Beginning in the late 1950s, engineers began utilizing deep foundation designs (i.e. drilled pier foundations) to reduce the risk of soils-related damage. Some of the first states to implement these designs included California, Colorado, and Texas. For the next three decades, the science and practice of foundation design changed little. In the late 1980s and 1990s, methodologies for predicting slab heave, pier lengths, and pier heave were published. This guidance helped provide engineers with the design basis that is commonly used today.

As new methodologies were published, the depth of piers utilized in construction became deeper over time. In Colorado, relatively significant increases in pier depth occurred around 1990. Professional organizations, such as the Colorado Association of Geotechnical Engineers, documented increasingly conservative (i.e., deeper and/or longer piers) designs over time; however, the cause for these design changes was questionable. More recent publications suggest that the mechanics of soils movement and pier behavior are more complicated than initially contemplated in the 1950s. Although design improvements have occurred with advances in science, evidence suggests that it is likely that litigation has had a more significant impact on design than engineering research alone.

Over the past 25 years, the western United States has seen a relatively significant increase in the amount of construction defect litigation. The timing of such litigation correlates with increases in pier length used in the design of new construction. While practical experience and continuing research has affected the design of deep foundations, data and testimony suggest that significant changes in foundation design may correlate with an increase in construction defect litigation.

A history of the progressive nature of foundation design will be presented. This presentation will also document advances in engineering design methodology and trends in foundation design over time.

Engineering Design, Construction Defect, Geotechnical

C20 Evaluating Structural Damage in the Collapse of a Coastal Bulkhead System

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After attending this presentation, attendees will learn several factors determining the assessment of damage to a site

containment structure, and how engineering principles were applied in a case involving the collapse of a bulkhead wall system containing a bayside residential building complex.

This presentation will impact the forensic science community by educating the public on some considerations engineers use to evaluate the damage caused by catastrophic events as well as differentiate it from pre-existing conditions in a building superstructure.

In this case, a 25-year-old site containment bulkhead collapsed at a seaside housing complex. The cause of the bulkhead collapse was determined to be an insufficient depth of the sheet piling system, which may have been intensified by scouring and an adjacent deepening of the bay navigation channel over several decades. Tidal variations and significant current velocities likely further saturated the retained fill, thereby increasing the hydrostatic pressure on the bulkhead, and eventually precipitating the collapse.

When the bulkhead wall collapsed, soil within the active pressure zone ruptured into the bay as well. One expert report claimed that the entire structure moved toward the water as a result of the soil movement, because the pilings inside the garage were all observed to be rotated at the top in the direction of the bay, as if to prevent the rest of the building from collapsing. No damage to the interior sheetrock finish was visible and/or documented, although it was alleged that the overall movement of the building complex may not have caused enough rotation to crack the finishes, even in the event of a structural collapse. Additionally, it is possible that the floor tiles may not have indicated visible cracking if the entire floor had moved together.

However, there were several factors that led to the different determination that the structural integrity of the building was not compromised by the bulkhead's collapse and that certain deficiencies were pre-existing.

First, the upper floors throughout the units did not contain significant cracking or lateral movement, which would be evidence of structural collapse or loss of building superstructure integrity. There was also no cracking observed in the hardened spray foam insulation in the garage and recent interior finish cosmetic work, which could be expected to show some displacement in the case of out-of-plane vertical displacement.

Second, the pilings that were observed to be rotated toward the site of the collapse were located farthest from the bay. If the collapse did cause the pilings to rotate, the pilings closest to the bay should have shown the most uniform rotation and the greatest lateral movement, which was not the case. The majority of the existing pilings were permanently set in two directions, and most of the pilings adjacent to the bay were actually measured to be laterally displaced away from the bay.

Finally, it was important to determine whether the observed structural conditions were due to a catastrophic event or related to original building as-built conditions. When constructing a residential structure, the initial process of connecting stringers to the pilings is a rough framing event with an inherent potential for rotation, so it is not unusual for wood pilings to be somewhat out of plumb. Specifically, if the rotation is random rather than uniform, this variation implicates an original as-built condition rather than one caused by a geotechnical failure event. Additionally, if the exterior sheathing is bowed and deformed around the existing piling butts above grade, this is similarly an indication that the pilings exist in an original as-built condition.

Overall, these factors led to the evaluation that the overall structural integrity of the superstructure was not compromised by the loss of the bulkhead in this case.

Bulkhead Collapse, Piling Rotation, Structural Integrity

C21 Air Conditioner Compressor Blasts Taking Lives in India: A Forensic Analysis

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After attending this presentation, attendees will have a better understanding of the various causes behind the compressor blast of air conditioners (ACs).

This presentation will impact the forensic science community by improving awareness of the fact that AC compressor blasts, though rare, are a harsh reality in a country like India. Recently, India has witnessed a surge in fatalities due to AC compressor blast. One important reason behind these incidences is non-adherence of safety guidelines by compressor manufacturers. Strict adherence to safety guidelines by manufacturer companies and engagement of well-trained, qualified professionals in AC repair jobs will definitely prevent such horrific incidences.

Worldwide, two types of household air conditioners are used: one is a window unit and the other is a split unit. The basic difference between the two is that a split AC has its compressor unit installed outside the room, whereas in a window AC, the entire assembly is installed at the window. Primarily, both consist of an evaporator, which receives the liquid refrigerant; a condenser, which facilitates heat transfer; one expansion valve, which regulates refrigerant flow into the evaporator; and one compressor, a pump that pressurizes refrigerant.

The focus of this presentation is on the workings of the compressor for the AC unit. The compressor is actually a large electric pump that pressurizes the refrigerant gas as part of the process of turning it back into a liquid. A typical AC compressor has two openings in the form of an inlet and an outlet (Figure 1).

The chief reason for AC compressor blast is extremely high pressure generated inadvertently by the use of dry nitrogen inside the compressor chamber while checking for leaks. Normally, AC compressors are capable of functioning at gauge pressure of about 1,000 pascals, but when it rises dangerously to 2,000 pascals, then a blast occurs. Most of the modern sophisticated compressors are equipped with safety valves which function to alleviate excess pressure. If the valve becomes dysfunctional due to a clogged pipe or if the compressor is not equipped with safety valves, then excess pressure causes the compressor blast.

In this instance, when the repairman reached the jewelry shop, he was allegedly unaware of the fact that the compressor had already been filled with dry nitrogen by another repairman prior to his visit. The moment he asked to switch on the AC unit, the blast occurred. Figure 2 shows the remnant of the compressor at the site. The blast was of such high intensity that a substantial part of his skull was blown off and all brain matter was discharged (Figure 3).

Another factor which may be responsible for such blasts is the use of oxygen instead of dry nitrogen to check leaks in the compressor. When oxygen combines with the compressor oil and a fire source is introduced, a compressor explosion occurs.

India is a developing country where ill or inadequately trained youths enter the AC repair market to earn a living, without knowing the potential hazards of their job. Only certified persons who take formal training from recognized institutes for AC repair should be hired. In addition, every compressor manufactured should comply with safety guidelines recommended by ANSI/AHRI

Standard 210/240 (Air Conditioning, Heating and Refrigeration Institute).

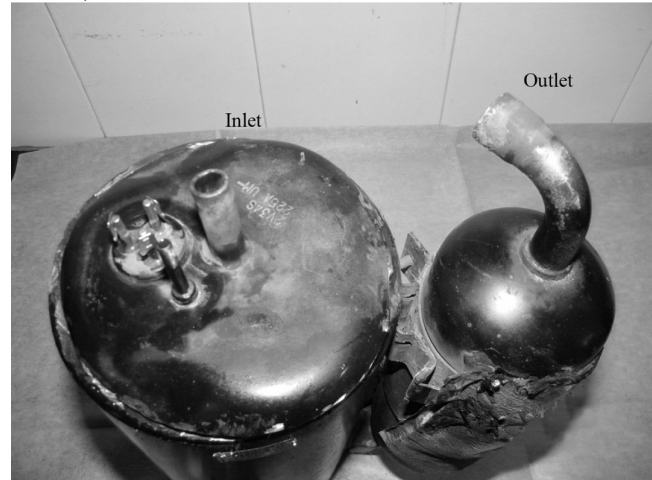


Figure 1: AC compressor showing inlet and outlet.



Fig. 2: Remnant of compressor (black in color) at blast site.



Fig. 3: Substantial part of skull was blown off and brain matter was discharged from the cranial cavity.

Air Conditioner, Blast, India

C22 Fuel-Control Valve Failure Analysis

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After attending this presentation, attendees will gain an understanding of readily-available tools and procedures used to answer the question of how a complex fuel-control valve can leak past some closed members but not others.

This presentation will impact the forensic science community by providing training on useful and inexpensive approaches to examine mechanical valve failures.

This presentation discusses the methods used to understand the failure conditions and mechanisms. These include bench tests, X-ray studies, and finally, the use of an inspection microscope.

Certain model trucks with dual fuel tanks use a fuel-pressure-controlled valve assembly to select from which tank to draw and return fuel. Under certain conditions, the fuel will be drawn from the rear tank, but returned to the front tank, leading to overfilling the front tank and eventually dumping fuel overboard.

There is a low-pressure (approximately 3 PSI) fuel pump in each tank and a high-pressure fuel pump on the frame rail, plumbed between the engine and the valve assembly.

The valve assembly, officially known as a "dual function reservoir," consists of a fuel inlet and return for each of two fuel tanks (front tank and rear tank), supply and return lines to the engine, a small fuel reservoir with fuel filter, and two over-pressure relief valves.

An O-ring-sealed, double-ended poppet valve is used to connect the engine fuel supply to either of the two fuel tanks. A second similar valve connects the engine fuel return to the appropriate tank return. These two valves are mechanically connected to force both to be actuated in the same direction at the same time, insuring fuel is drawn from, and returned to, the same tank.

During normal operation, the driver selects a tank from which to draw fuel with a switch on the vehicle's dash. This switch supplies power to the low-pressure fuel pump located in the desired tank (as well as connecting the correct fuel sender to the vehicle's fuel gauge). The in-tank fuel pump pressurizes the fuel line to the valve and causes a diaphragm attached to the valve linkage to move the valves to one end of travel, thus connecting one tank to the supply and return lines, and shutting off the connections to the other tank. When the driver selects the other fuel tank, the pressure on one side of the diaphragm is released, and the pressure on the other side increases, thus moving the valves to the opposite ends of their travel, and changing the tank connections.

In practice, sometimes the sealing of the return line to the front tank is incomplete, thus allowing fuel to return to the front tank when the rear tank is selected. If the front tank is nearly full, fuel can leak from the front tank and drip on the ground and vehicle frame.

There is a manufacturer-supplied bench test procedure to verify that the valve assembly is functioning correctly. A number of new and used valves were tested, and the failing valves were labeled for further study.

The valve assembly is able to be non-destructively disassembled, and when a failing valve is disassembled, sometimes there is no obvious defect — the O-rings appear undamaged and are in the correct location, and the other valve parts all appear to function properly. A method was needed to determine, in these cases, where the valve leak was occurring.

Using an industrial X-ray machine, it was determined that while the O-ring positions in the failing valves were different than normal valves, sealing should still occur.

Since valve sealing did not occur, examination under an inspection microscope was carried out, and it was determined that a combination of mechanical issues was preventing the O-rings from properly contacting the valve seat, thereby, allowing leakage.

Using these non-destructive testing methods, the failure mechanism of the failing valve assemblies was isolated, duplicated, and proven.

Valve, Failure, Analysis

C23 *In Situ* Hardening of a Steel Tank: Carbon Diffusion Over 35 Years at Ambient Temperatures

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After attending this presentation, attendees will understand the importance of diffusion in materials, particularly in surface hardening of steel.

This presentation will impact the forensic science community by presenting a case study where low temperature diffusion has a large impact on the properties of steel.

During recoating of the interior of a 35-year-old, 1.2MG municipal water tank, the interior coal tar pitch epoxy coating was being removed by sandblasting where the goal was a white metal finish. Initial sandblasting with silica resulted in unexpectedly low removal rates. Time was of the essence, so the abrasive was switched to steel abrasive, a more expensive material. The blasting still took much longer than scheduled and anticipated.

A disk taken from the roof of the tank was cut in half; Piece A, not cleaned in any way, and Piece B, sweep blasted with steel abrasive. Optical microscopy, Scanning Electron Microscopy (SEM), and Energy Dispersive X-ray Spectroscopy (EDS) were performed on both A and B.

Uncleaned A, under the optical microscope (Figure 1), showed a cracked coal tar pitch epoxy coating. In some areas, the coating had spalled away and red iron oxide scale formed. Under SEM, a region containing both coating and corrosion was examined (Figure 2). The corrosion had a typical iron oxide appearance (Figure 3). The coal tar coating (Figure 4) showed some surface deposits and surface cracks.

Elemental analysis of Piece A by EDS was unremarkable, with carbon, iron, and oxygen from the coal tar coating and steel tank material. Silicon, aluminum, potassium, magnesium, calcium, sulfur, and chlorine were also present, likely deposits from the well water held in the tank.

Optical microscopy of B showed that the sweep blasting, while incomplete, had removed most of the coal tar coating (Figure 4). Residual red iron oxide and black regions consistent with the coal tar coating seen on the uncleaned piece were found. Piece B was examined in the SEM. The surface was somewhat rough, indicating incomplete sweep blasting. EDS of the area again revealed no unexpected elements.

Spot elemental analysis was performed on an area of sweep-blasted B, where a cross-section from the pitch coating to the bare steel was visible (Figure 6) at three locations; pitch coating, mid coating, and bare steel (Table 1.). The additional elements detected, likely deposits from groundwater, were silicon, calcium, aluminum, magnesium, potassium, sulfur, and chlorine.

Element	Atomic %		
	Spot 1	Spot 2	Spot 3
Carbon	50.1	33.2	1.7
Oxygen	34.9	31.8	22.6
Iron	0.4	30.1	65.3
Additional elements	14.6	4.9	10.4

Table 1. Concentration of elements found at three different spots on a cross-section of Piece B.

At Spot 1, the top of the coating, carbon from the coal tar pitch epoxy was detected. Hydrogen was also likely present, although EDS cannot detect it. At Spot 2, a significant amount of iron was detected. The ratio of iron to oxygen was approximately 1:1, which suggests iron oxide of the form FeO, a black oxide. The presence of carbon suggests carbon diffusion into the iron oxide. At Spot 3, the primary element present was iron.

Piece A is representative of the present condition of this 35-year-old tank and coal tar coating. Over time, volatile organic compounds evaporated from the pitch, causing it to lose elasticity. Cracks in the coating then initiated and grew. Water penetrated through the cracks to the steel and formed ferrous oxide (FeO), the iron corrosion that forms in limited-oxygen environments. The corrosion propagated underneath the coating (Figure 7), allowing spallation. The iron, then exposed to oxygen, converted from black FeO to red Fe₂O₃.

Piece B reveals a second process, carbon diffusion. Carbon will readily diffuse into iron, at a rate dependent on temperature. The carbon gradient discovered in Piece B indicates that the carbon diffusion process has been taking place over the 35-year lifetime of the tank. Although the maximum ambient temperature was relatively low (~100° F), the long time allowed significant carbon diffusion to occur, causing surface hardening of the low carbon steel. This diffusion rate was verified by Fick's Second Law.

The goal of sand blasting was to remove the pitch layer and the top surface of the steel, leaving a clean steel surface for recoating. The increase in hardness from that of the expected low carbon steel material, to a high carbon steel of around 2.5%, resulted in exceptionally low material removal rates by sand blasting.

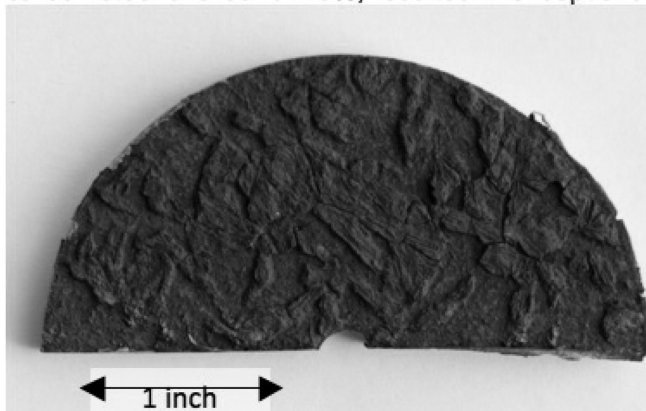


Figure 1. Water tank specimen, Piece A, no cleaning.

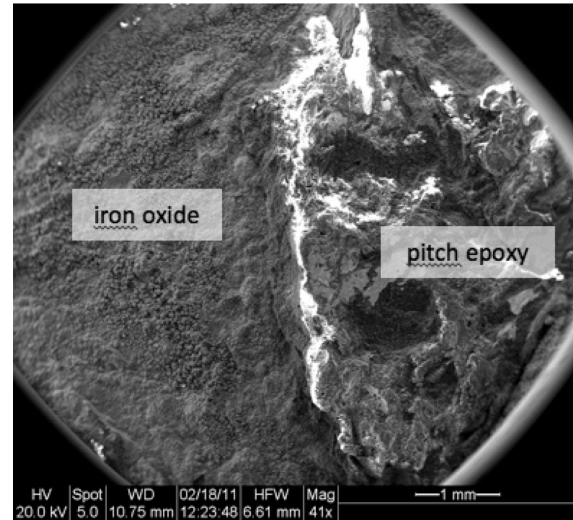


Figure 2. Electron micrograph of A, in a region with both red iron oxide corrosion and pitch epoxy coating.

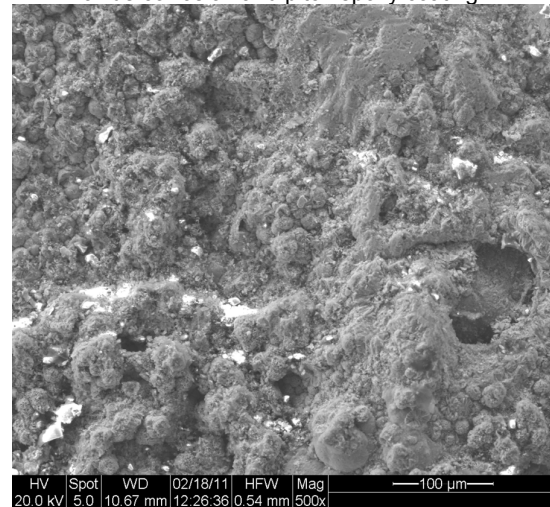


Figure 3. Electron micrograph of corrosion from A. The bubbles are typical of rust; EDS analysis confirmed the composition of the rust to be Fe₂O₃.

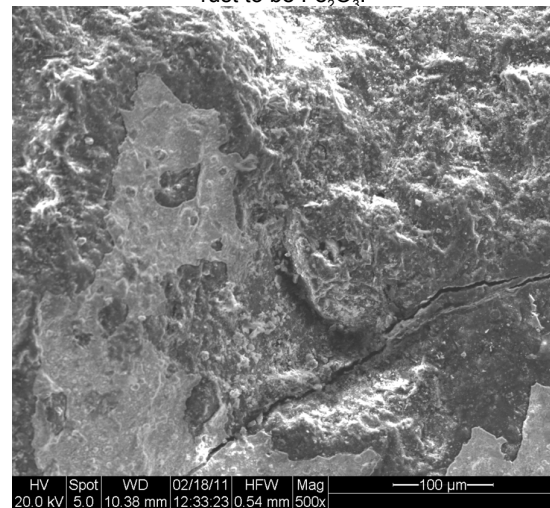


Figure 4. Electron micrograph of the pitch epoxy coating from A. The light areas are surface deposits. The crack is typical of those found in the coating.

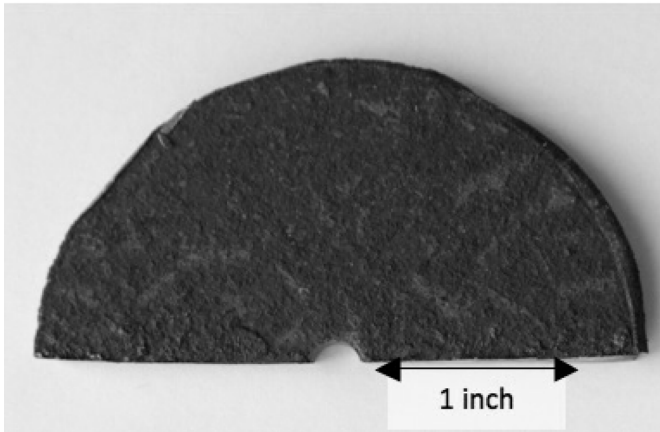


Figure 4. Water tank specimen, Piece B, sweep blasted.

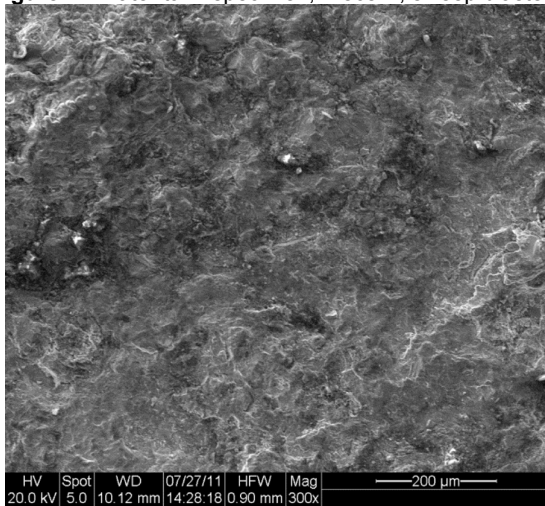


Figure 5. Electron micrograph of B. The rough surface is a result of sweep blasting with steel abrasive, which did not penetrate to the steel substrate.

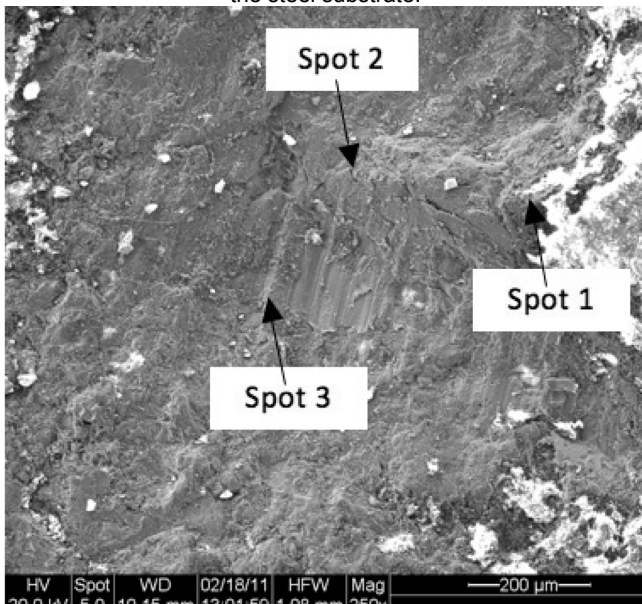


Figure 6. Area of EDS analysis.

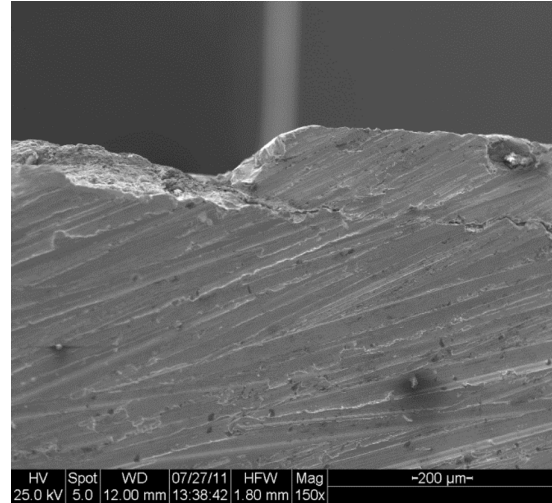


Figure 7. This cross section shows the propagation of corrosion beneath the coating.

Diffusion of Carbon in Steel, Corrosion, Oxidation

C24 Crashworthiness Investigation of Modern Sedan Bumper Designs

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WITHDRAWN

Bumper, Crashworthiness, Composite

C25 Is Fracking the Key to Americas Achieving Energy Independence Via Natural Gas Production?

Peter Alexander, PhD*, 23595 E Irish Place, Aurora, CO 80016

The goal of this presentation is to educate attendees regarding energy production and forensics needs.

This presentation will impact the forensic science community by providing education concerning fracking and other natural gas resources issues.

America's need for oil from the Middle East has driven U.S. foreign policy for generations and has benchmarked the price we pay for domestic energy. Suppose that over the next decade the U.S. could become energy self sufficient or a net exporter of energy. Such an outcome could reconfigure our national security and foreign policies. It could also produce a revolution in our ability to restore the U.S. as a manufacturing powerhouse fueled by low cost energy. Natural gas prices, which have ranged from \$2 to \$12 per thousand cubic feet over the last decade, are currently \$3 to \$4 in the U.S. versus \$12 in Europe. This low current domestic price is largely due to the increased production of shale gas.

What has changed to suddenly make such previously outlandish outcomes possible? The answer is that advances in directional drilling and the application of a long-standing technique called hydraulic fracturing or "fracking" have enabled U.S energy

production companies to tap and produce vast quantities of natural gas and oil resident in massive subsurface shale formations located in various parts of this country which are economically unproductive without the application of fracking. There are at least ten regions in the U.S. in which “shale plays” are currently active. These include the Bakken formation in the Dakotas and the Marcellus formation along the East Coast.

In fracking, a vertical well is drilled, usually thousands of feet below the level of any aquifers present. The well is encased by a steel pipe, which is further encased by cement. Fluid is pumped at high pressure into the well and is conveyed to the shale formation where it opens avenues for oil and gas removal. The fractures are kept open by solids (sand) pumped in with the fracking fluid. Directional drilling allows one drill pad to gather oil and gas from many directions using one vertical well, making the process more economically attractive.

According to the U.S. Energy Information Administration, within 25 years shale gas production is slated to equal or exceed natural gas production from all other sources combined. Annual U.S. natural gas production in this country is estimated to reach 33 trillion cubic feet by 2040, up from 23 trillion cubic feet in 2011. U.S. natural gas production is not expected to plateau until 2040.

As with any new application of previously proven technology, there are risks as well as rewards associated with this opportunity. Some of these risks are detailed below:

- Fracking requires large quantities of water. This raises questions regarding water availability, particularly in arid regions. In the West, the water from the Colorado River is very close to being oversubscribed. The fracking fluids can carry contaminants from the subsurface rock matrix back to the surface. Can the water used be cleaned up and reused? Will fracking contaminate the ground water? How will the vast quantities of fracking fluids be disposed of?
- Gas emissions into the atmosphere, from drilling and fracking operations, are of concern. These emissions can include methane, radon, and other gases.
- In populated regions, truck traffic, noise, smells, the presence of unsightly operational hardware such as pumping stations and storage tanks, and the loss of privacy can stimulate negative reactions from neighbors.
- Numerous legal and regulatory issues manifest themselves. In many states the mineral rights are treated as a separate entity, not the property of the homeowner. Multiple regulatory organizations are vying to regulate the use of fracking. These include homeowner groups, cities, states, and the federal government.
- Economic issues are critical. How much natural gas resource is available at what extraction and transport cost?

For most of the above problems there are solutions, but a paramount question is: can the public trust that the solutions will be properly applied? A number of the above topics will be addressed by the speakers that follow.

Forensics fits into this picture when there is a problem or a mishap. The forensics discipline needed might be in the area of geology, engineering, environmental, or some other area.

Fracking, Energy, Gas

C26 Minimizing Risks From Natural Gas Production, Distribution, and Use

Drew S. Nelson, MS, 301 Congress Avenue, Austin, TX 78705*

After attending this presentation, attendees will have a better understanding of some key environmental risks to water, air, and climate associated with shale gas development in the United States, and some of the regulations and best practices that can help minimize these risks.

This presentation will impact the forensic science community by providing a broad overview of the trade-offs between the benefits of increased natural gas development and the very real risks to public health and the environment.

New supplies of domestic natural gas have caused a drop in price that has benefited consumers and the environment alike. Low-cost natural gas is one reason why proposals for new coal-fired power plants have been withdrawn across the country and why old, inefficient, highly polluting coal plants are finally retiring. While there is no question that natural gas burns cleaner and has roughly half the carbon content of coal, natural gas has its own challenges. Producing new supplies of natural gas entails some fairly significant risks to public health and the environment, and natural gas can have a substantial greenhouse gas footprint in its own right.

Minimizing environmental risks related to natural gas production, distribution, and use is essential. This presentation will outline some of the key environmental risks associated with the increase in natural gas production and how they can be reduced.

Water: Oil and gas drilling creates multiple potential pathways for water contamination, including surface spills, improper well construction, and improper disposal of waste water. At the well site, groundwater can be contaminated through faulty well construction or chemical spills at the surface. Poor well casing or cement jobs can increase the chance of methane leaks or groundwater contamination. Groundwater can also be contaminated by mishandling waste water. Water used in the process of hydraulic fracturing, as well as water released from the shale along with the gas, must be treated and disposed of properly. If fracking wastewater is mishandled, it can contaminate surface and groundwater.

Air Pollution: Air pollution from gas production can be significant. A study conducted by Southern Methodist University in neighborhoods around Dallas concluded that air pollution produced by the drilling, production, and delivery of oil and gas is equal to the pollution produced by four million cars and trucks driving daily in the Dallas-Fort Worth area. In parts of Wyoming where gas production occurs, once pristine air now fails to meet federal health standards.

Climate: Natural gas is primarily methane — a powerful greenhouse gas many times more potent than carbon dioxide; therefore, even small leaks can rapidly accelerate global warming. No one is sure exactly where the methane leaks and releases are or how much escapes. Estimates range from 1-8% of total production. A number of companies and research institutes are working to gather data to better define the leak rate. If leak rates are high, and are not reduced, methane emissions could undermine the greenhouse gas advantage natural gas offers and actually make things worse for the climate system.

Strong regulations can help mitigate some of these concerns and are being developed and implemented. For example, we have seen an increase in the number of states that mandate disclosure of chemicals used in the hydraulic fracturing process, from 3 to 20 over the last three years. These states represent most of the major oil-and-gas producing states, and are governed by both Republicans and Democrats, showing that strong, bipartisan, and effective regulations can be crafted on these issues.

Natural Gas, Hydraulic Fracturing, Risk

C27 The Macro Implications of Shale Gas: U.S. Liquefied Natural Gas (LNG) Exports and Other Margins of Response

Kenneth B. Medlock III, PhD, Rice University, Baker Institute, MS40, 6100 Main Street, Houston, TX 77005*

After attending this presentation, attendees will gain insights into the broader economic and geopolitical implications of the rapid expansion of shale gas production facilitated by upstream innovations in the recent past.

The presentation will impact the forensic science community by providing a backdrop and context for the shale gas resource and what it means.

A decade ago, market players were making large capital investments to facilitate the import of Liquefied Natural Gas (LNG) to the United States. However, innovations involving hydraulic fracturing and horizontal drilling led to the dramatic growth of domestic gas production from shale. Now, the U.S. is considered a possible exporter of LNG. Importantly, the commercial aspirations of firms that seek to seize the apparent profit opportunity offered by exports run headlong into concerns that allowing exports from the U.S. will force prices up and drive increased volatility, thereby negatively impacting industrial activity and household budgets. Hence, the issue of allowing LNG exports from the U.S. has entered the political realm. Several studies have assessed the impact of U.S. exports on domestic prices. Most of these studies generally assume LNG export volumes to be exogenous and, therefore, not allowing interaction between domestic and international markets to influence the volume of trade. This presentation will present a basic international trade framework to argue that: (1) the impact on U.S. domestic prices will not be large if exports are allowed; (2) the long-term volume of exports from the U.S. will not likely be large given market developments abroad; (3) there will not be "one price" for natural gas as the cost of trade will dictate regional pricing differentials; (4) concerns about U.S. domestic price volatility increasing if exports commence are unfounded; and, (5) the impacts of simply allowing trade to occur unfettered will be transformative to markets abroad.

Shale Gas, Natural Resource Economics, International Trade

C28 Pipe Joint Failure Analysis at an Oil Field Fracking Site

Marc A. Meadows, BS, PE, Meadows Analysis & Design, 4150 International Plaza, Ste 850, Fort Worth, TX 76109*

After attending this presentation, attendees will understand the dangers of temporary pipes used in the fracking of oil and gas wells.

This presentation will impact the forensic science community by bringing to light the dangers of operating around temporary piping while servicing oil and gas wells.

Servicing a gas or oil well involves installing temporary piping. This piping is used to transport water, chemicals, sand, and other materials around the well site and into the well for many actions including fracking. The pressures can be on the high side, in fact up to 15,000psi. The pipe, therefore, is designed especially for this type of service. Normal pipe with threaded fittings, while rated up to 2,000psi, is not designed for the repeated application of high pressure. The type of pipe used most often in the oil and gas industry is called "1502" and each link and fitting is serial numbered. The pipe is joined with hammer fittings and the threaded joint does not come in contact with the pressure. Some service companies test this pipe as often as monthly to make sure it is in good shape. Using this pipe to transport abrasive and corrosive materials wears it out from the inside. Certified inspections are a must and include pressure testing. Sometimes the pipe still fails and the resulting accident can be devastating to personnel and equipment. The testing is done in bunker-style units to protect surrounding equipment and personnel. This pipe necessarily has a thick wall and therefore is very heavy. A failure of a joint or section of pipe releases the transported material at extreme pressure. The most dangerous situation involves an energized fluid. This term defines a material that has a large gas component that is pressurized. The expanding gas escaping from a failure will cause the pipe string to disrupt, fly apart, damage adjoining equipment, or harm personnel. A non-energized fluid is not as violent, but if pumps are running to maintain volume, up to 100 barrels of fluid per minute could exit the failure! Although there are restraint systems available, not all operators are required to use them.

A case study of a well service employee who straddled a pipe while turning a valve with a wrench will be presented. The torque from the wrench initiated a failure of the joint. Since the joint was under pressure, it violently separated, throwing the employee and his helper many feet. The employee suffered significant injuries to his leg. Lawsuits were filed and an expert was retained by the plaintiff's attorney. Nothing happened for many months, but the case became active when a blood clot, possibly from the injured leg, caused the man to die.

This presentation will illustrate the dangers of the temporary pipe, what went wrong in the case study, and what can be done to reduce the incidents of injury due to this necessary action.

Fracking, Oil, Piping

C29 Tour Bus Limit Handling Degradation With Suspension Modification

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The goal of this presentation is to show the results of how modification to bus suspension affects controllability.

This presentation will impact the forensic science community by defining the definition of bus limit handling with and without suspension modifications.

A 29-passenger tour bus suspension was modified to increase the ride comfort. The rear leaf spring normally has a steel shackle for rear attachment. The shackle was replaced by a stack of rubberized material that would not only support the spring, but would absorb some of the road vibration. Another result of the modification is that the rear of the rear leaf spring could also have up to an inch of additional lateral motion. The rear spring support loses some of its stiffness. The additional lateral motion of the rear

axle during a turn, particularly at higher turning rates, can reduce the controllability of the overall bus.

A series of track tests were performed to evaluate and quantify the results of the suspension modification. The bus was fully instrumented to measure both control inputs and dynamic outputs. The bus was equipped with outriggers for safety. Movie cameras, both onboard and off, documented the tests. The vehicle was equipped with a controller that could be programmed to input steering. Braking and throttle was controlled by the driver. The measurements included 3-axis linear acceleration, 3-axis angular acceleration, velocity, steer input, braking input, and throttle input, as well as ride height at each wheel and rear axle lateral location.

To evaluate the controllability of the vehicle in near limit turning, two basic tests were performed. A modified Federal Motor Vehicle Safety Standard (FMVSS) 126 and J turn. The FMVSS 126 is a standard to evaluate the effectiveness of the directional stability control for vehicles. While this bus was above the weight limit for direct application of this standard, it was useful for turning evaluation. The basic standard requires some preliminary warm up and tire break in tests. The turning is basically a modified sine wave with a dwell on the second portion of the turn when the steering is at its maximum. The J turns are simply a turn in one direction. The severity of the turns is increased until a limit is reached.

Side slip in vehicle dynamics is the angle between the heading of the vehicle and the direction of travel. In addition to the traditional performance criteria of FMVSS 126, the duration and magnitude of the side slip was a measure of control performance. FMVSS 126 has as criteria the return-of-Yaw rate after the steer input at milestone times.

The attachment of a lateral bar from the frame to the axle (panhard rod) is designed to limit the lateral rear axle motion. The vehicle was also tested with a panhard rod attached in the case of the rubberized modification.

Tests were conducted on a production suspension with a steel shackle, the vehicle with rubber support at the rear, and the vehicle with the rear rubber supports and a panhard rod.

The bus was most controllable and passed all the FMVSS 126 criteria when in the original design configuration with the steel shackle. The addition of the rubberized rear attachment significantly reduced the limit turning performance of the vehicle with failure of the FMVSS 126. The panhard rod improved the performance of the rubberized suspension with some of the FMVSS 126 criteria passing.

The conclusion is that rubberized suspension that allows approximately one inch of lateral motion of the rear of the rear leaf spring support can lead to degraded limit turning performance.

Bus Limit Performance, Bus Track Tests, Bus Handling Testing

C30 The Cause Analysis of a Farm Tractor and Motor Grader Collision Fatality: A Case Study

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The goal of this presentation is to share knowledge with the scientific community by thoroughly examining the case information and presenting the details of the collision analysis.

This presentation will impact the forensic science community by proving case background and demonstrating the analysis used to determine the cause of collision.

A compact 1,900lb utility tractor commonly used in vineyard service was impacted by a 19,500lb motor grader. The tractor operator was ejected and killed. A forensic analysis was performed to determine the cause of the collision.



The incident occurred during the vineyard's canopy maintenance season that includes replacing the old, non-producing plants as needed. The tractor was following the planting crews while pulling a water storage trailer that allowed the operator to water each new plant by hand. At the end of the row, the tractor entered a service road between the blocks and was completing a U-turn in order to re-enter the block between another pair of rows.

It is unknown if the tractor operator was wearing full-coverage ear protection at the time, though it is unlikely this operator was aware that the grader had previously graded service roads at an adjacent site and was now in transition to another location. The grader's course on the service road intersected that of the tractor while negotiating its U-turn.



A late attempt at collision avoidance was initiated by the grader operator attempting to steer around the tractor. The collision knocked the tractor over onto its left side, and the operator was ejected. Physical evidence shows that the tractor operator was not wearing a lap belt. The grader's left front wheel mounted and rolled over the tractor, then over the operator, causing fatal head injuries. The grader continued down a slight decline for another 140ft after the operator dropped the grading blade as a means to stop the grader whose engine had apparently stalled.



A visibility study was undertaken to determine the sight lines of each operator. The eye-level of the tractor operator was approximately 5.7ft above grade, and the top of the Rollover Protection Structure (ROPS) is 6.5ft above grade. The tractor's orange safety flag rose about another 5-6ft above the ROPS. The vine trellis top wire height adjacent to the tractor's path was approximately 4.8ft, with the top of the vine canopy at about 2.5ft above grade. The eye-level of the grader operator was approximately 8.8ft above grade, and the top of the grader's cab is 11ft above grade. Despite a gentle rise in the service road prior to the point of collision, the grader operator could begin to observe the tractor's orange safety flag approximately 350ft before impact. Therefore, it was concluded that a visibility impairment was not a cause of the collision.

A demonstration video using an exemplar tractor shows the time to complete a U-turn and attain an approximate pre-impact orientation is approximately 6.8 to 7.7sec. In the video, the tractor's flag and operator's head are clearly visible above the vine canopy height.

Three series of tests were conducted on the subject grader using a Logitech® V-Box II Lite to measure distance, speed, and deceleration. In the first series, the grader's operational speed for each of its six forward gears was determined. The grader's top speed in 5th gear (measured on a level dirt road) was approximately 17.5mph. In the second series, the average deceleration using

only the service brake was measured. Pre-braking speeds of 10.7 to 12.4mph (4th gear) yielded average deceleration rates of 0.177 to 0.234g, whereas pre-braking speeds of 16.3 to 17.2mph (5th gear) resulted in average deceleration rates of 0.186 to 0.232g.

A third test series was designed to measure the stopping distance and deceleration rate by only lowering the blade as a means to stop the grader. Three tests conducted at approximately 11mph demonstrated stopping distances of 12.3, 8.5, and 11.8ft, respectively. The average deceleration for these tests was 0.154, 0.148, and 0.176g, respectively. A fourth test conducted at 17.3mph, resulted in a stopping distance of about 23.8ft, with an average deceleration of 0.262g.



A time-position analysis of the grader at 17mph showed it was approximately 14sec from impact when the tractor was reasonably expected to be in view. Assuming a 2sec perception/reaction time, and allowing 2sec for the grading blade to make soil contact, the grader operator could have stopped in about 125ft. Therefore, it was argued that the grader operator had failed to use precious time available to avoid the collision.

Farm Tractor, Motor Grader, Fatality

C31 Swaying Trailer Caused a Fatal Crash: How Can That Happen?

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After attending this presentation, attendees will learn the conditions and parameters involved in causing a trailer under tow to become unstable and sway, resulting in loss of driver control leading to a fatal crash. Attendees will also learn how a published basic mathematical model was modified and expanded to account for additional real-world phenomena that are present in the actual case.

This presentation will impact the forensic science community by informing those who are reconstructing crashes involving trailer sway. Certain tow vehicle-trailer system design parameters can lead to unstable operation in response to interactions at tire contact patches on road surfaces.

A suburban family began a journey from their home for a Memorial Day weekend of camping. The man of the house was driving a Sports Utility Vehicle (SUV) with his wife as a front seat passenger, his children and pet dogs as rear seat passengers, and camping gear in the rear storage area. He was towing a travel trailer with modest living quarters, camping gear in the interior,

and externally mounted bicycles on the back trailer end. The loaded trailer weight was approximately 75% that of the loaded SUV weight. Since the trailer tongue weight is sufficiently heavy, a distributive hitch was used for the purpose of shifting some of the tongue weight forward from the SUV rear axle onto its front axle. Before starting the trip, a quick check of trailer brakes showed them to be mechanically functioning; however, brake performance was not thoroughly tested and no brake adjustments were made.

A few miles after the entrance ramp onto the interstate highway, they entered a road construction zone wherein temporary jersey barriers separated the opposing travel lanes. The construction zone began a few hundred feet beyond a moderate hill crest with a moderate right curve. Temporary travel lanes were offset and consumed most of the median space. The left lane for the SUV driver was concrete and the right lane was asphalt about 1-2 inches lower in elevation. The ridge between the two lanes followed within a few inches from the lane divider line.

While approaching the hill crest preceding the construction zone, the trailer began to sway in yaw that a following driver described as side-to-side. The swaying stopped and normal operation resumed for a short distance. Beyond the crest, the trailer began swaying unstably with increasing amplitude of oscillation. The SUV driver actions did not stop the swaying which caused him to lose control. The SUV front impacted and climbed the temporary median jersey barriers. The catapulted SUV front right wheel came down onto the hood top of an oncoming vehicle in the opposite travel lane nearest the barrier. The oncoming vehicle driver and rear seat passenger behind him suffered fatal head injuries as the roof was peeled back.

Multiple defendants with varying assets and insurance limits were named in the ensuing legal action. An expert was retained to evaluate parameters and variables involved in causation of trailer sway. This was deemed necessary to assess the SUV driver's contributory negligence for the unstable operation which culminated in trailer sway, loss of vehicle control, and the fatal crash.

The subject of trailer sway and instability is addressed by Hans B. Pacejka in the 2006 SAE publication entitled, *Tire and Vehicle Dynamics* and perhaps in other references. A mathematical model is presented consisting of a set of linear differential equations with constant coefficients including several design parameters. Questions were posed about contributions of several parameters known to exist for the subject case that are not included in the published mathematical model. The modified and expanded published model was to include these additional parameters. The expanded model was programmed for computer computations and stability determinations based on classical control theory criteria. Calculated results are presented demonstrating effects of various system parameters. Questions are posed about causation contributions of the SUV driver, the SUV, trailer, hitch manufacturers, and the dealers.

Accident, Trailer, Sway

C32 Wrong Way DUI Head-On Collision Reconstruction Using DashCam Video

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The goal of this presentation is to share knowledge with the forensic community by detailing the analysis and reconstruction of a fatal collision using the video camera images recorded by an onboard law enforcement camera.

This presentation will impact the forensic science community by virtue of the severity of the impact being analyzed and

demonstrating tools which are effective in collision reconstruction.

A family of six, in two vehicles was heading home after an evening out of town. A driver and front passenger in a small van were being followed by a passenger vehicle containing the driver, front passenger, and two rear-seated occupants. Both vehicles were traveling in southbound lane #2 of a two-lane highway with a posted speed limit of 65mph.

An intoxicated individual entered the freeway south of the two-vehicle caravan, and began to drive the wrong way in a mid-size Sports Utility Vehicle (SUV). This operator, under the influence of alcohol, cocaine, and/or other drugs, was driving northbound in lane #1.

From a location between the three converging vehicles, a law enforcement officer responded to the call of a wrong-way driver and entered the southbound traffic lanes from an adjacent on-ramp. Without activating the overhead emergency lights, the officer began to merge into lane #2 at a speed about half that of the caravan approaching from the rear, also in lane #2. The lead vehicle of the caravan was forced to merge into lane #1 to avoid a collision with the rear of the officer's vehicle, thereby becoming aligned with the path of the approaching SUV. The second caravan vehicle followed the first vehicle into lane #1.

Puzzled by the strange encroachment of the officer's car, the lead vehicle operator's attention was momentarily distracted. Only after this vehicle passed the officer's car were the overhead emergency lights activated, approximately 7sec before impact. The lead vehicle operator, thinking they were being pulled over, activated the turn signal and began to merge toward the right-hand shoulder. In doing so, they narrowly missed impacting the wrong-way SUV, but the SUV then slammed head-on into the passenger car.

The impact killed both front seat occupants of the passenger car and seriously injured the two rear-seated passengers, all of whom were wearing their seat belts. The wrong-way driver escaped with only minor injuries.

The video camera, secured to the roof header inside the officer's vehicle, is triggered to begin recording upon activation of the overhead lights. Additionally, the onboard system also records one minute of footage prior to this trigger. The video image captured the offset head-on collision having a closing speed in excess of 120mph. This video was used to determine the pre-impact collision events.

A freeway section north of the point of impact was surveyed with a robotic Total Station device, and a scale diagram was prepared by a licensed professional land surveyor. Using known locations of the lane lines and artifacts observed in the video images, a time-position history analysis was used to determine the pre-impact speeds of the three southbound vehicles. It was determined that the officer's vehicle began its merge into southbound lane #2 at approximately 32mph. The van in lane #1 passed the officer at approximately 60mph. Next, the passenger car that was following the van at a distance of approximately 160ft, passed the officer's vehicle also at approximately 60mph.

The approach trajectory of the officer's vehicle descending the on-ramp until merging into lane #1 was compared to the officer's potential view behind in the direction of the approaching caravan. The pre-impact positions of the caravan were likewise considered. It was declared that the officer was not aware of the caravan until being passed, but the officer had approximately 18sec of time to observe the southbound vehicles and activate the overhead lights with the potential to warn of the immediate collision hazard ahead.

DashCam, Wrong Way DUI, Collision Reconstruction

C33 Evaluation of Detection Systems for Large Mobile Equipment

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After attending this presentation, attendees will understand methods that operators of mobile equipment can use to detect and identify objects or obstructions proximate to their machines in order to avoid contacting them when the machines are in motion.

This presentation will impact the forensic science community by describing the methodology used to test detection systems as well as the results, and by promoting dialogue and discussion regarding the efficacy of using detection systems on large mobile equipment with areas around them outside of the operator's normal field of view (i.e., blind spots).

Three different detection systems were evaluated for their ability to either sense objects and alert the operator or allow the operator to perform a timely visual assessment of the objects' proximity to the machine: one that was radar-based; a camera with a monitor; and, a parabolic mirror. For the purpose of this research, an excavator was selected because of the large field lateral to and aft of the cab and engine (house) that was not readily viewable by the operator. Each system was evaluated with the use of a 20ft by 30ft grid.

The research study provided a basis for analysis of the field of view/detection of various systems. In reviewing the line-of-sight testing methodology in SAE J1091 and ISO 5006, it was determined that similar evaluations of the mirror, camera and proximity sensor required modification, because the near field testing performed was different than the 12-meter threshold used by the SAE and ISO tests. Quantification and repeatability of the tests were enabled by using a grid system behind the excavator. The grid was a 12 inch by 12 inch matrix. The furthest aft center point on the excavator was the machine reference, and a plumb bob dropped from that point was used as the origin for the X and Y axes on the grid.

The proximity sensor consisted of an alarm attached to the detector via a cable between the rear of the machine and the cab. The alarm used colored lights and varying beep frequency to warn the operator how far an object was within the detection zone. The detector was affixed to the aft end of the excavator and located 52in. Above Ground Level (AGL). A person approached until the alarm began to emit beeps. The corresponding location of the detection threshold on the grid was then documented.

The camera system consisted of a waterproof camera and black and white LCD monitor. It was mounted to the top of the aft end of the excavator at a height of 85 inches and angled downward so that the bottom of the image in the monitor corresponded to the edge of the excavator tracks. Prior to testing, it was decided that determination of the visual field would be based on the threshold of 36 inches AGL. A subsequent series of tests were performed to quantify the range of the visual field as part of the error rate analysis. The same methodology was used. The lower boundary of the visual field was determined based on a threshold of 4 inches AGL.

The mirror system was a convex parabolic mirror with an effective diameter of 10 inches, mounted on a curved rod with a base intended to mount on the house. With the operator sitting in his seat and looking over his right shoulder, the mirror was located within his line of sight and angled to maximize his view of the area of the rear of the machine. The center of the mirror was 93 inches. AGL and positioned in the "passenger-side" rear quadrant of the house. Prior to testing, it was determined that either a 36-inch-tall object or "that's a person" would be the threshold for establishing the presence of objects.

Detection patterns from the tests of the three systems are

presented with an error rate analysis.

Although the efficacy of the systems may be cause for debate, they can assist an operator of mobile equipment in detecting objects proximate to the machine. They work best when used in combination. Obstructions or objects in the blind spot would cause the proximity sensor to alert the operator, who would then confirm their presence visually using the mirror or looking at the camera monitor.

Detection Systems, Blind Spots, Machine Safety

C34 Electrochemical Detection of Explosives and Their Metabolites in Fingerprint Sweat Using Molecularly-Imprinted Polyaniline Films

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After attending this presentation, attendees will understand the potential of molecularly imprinted electroactive polymers for the production of sensitive and specific biosensors for the detection of illicit excreted substances.

This presentation will impact the forensic science community by demonstrating the potential of molecularly imprinted electroactive polymers to be used for the electrochemical detection of trace amounts of explosive metabolites in the fingerprint sweat of suspected bomb makers. The capabilities of the methods presented herein may also be adapted to the detection of drugs and their metabolites. This presentation will also impact the field of biomedicine by potentially providing novel biosensors for the detection of specific biomarkers in sweat to diagnose disease.

The illegal production and transportation of explosives poses a constant threat to public safety and national security. Those individuals involved in the production and transportation processes are routinely exposed to these compounds, which are inevitably absorbed into the body, metabolized, and subsequently excreted through matrices, such as sweat. A method is proposed for the detection of these excreted explosives and their metabolites within the sweat of a fingerprint. Due to the minute volumes of sweat and the trace concentrations of parent compound and metabolites therein, this method requires both high specificity and sensitivity, while simultaneously being cost effective. Electrochemical devices are generally considered the most appropriate to satisfy those goals. The electroactive properties of polyaniline (PANI), as well as its ability to be molecularly imprinted when synthesized in the presence of a template molecule, make PANI an ideal polymer for this proof of principle. Subsequent removal of the template molecule leaves a PANI film with pores that are specific in size, shape, and polarity to the target molecule. Alternate methods of coating and imprinting of PANI nanofibers are being investigated as a means of imparting higher sensitivity to these films, owing to the greater surface-to-volume ratio offered by the nanofibers. Detection will be achieved by oxidizing/reducing the target molecules adsorbed on the imprinted PANI film, by use of cyclic voltammetry. The feasibility of this concept will be investigated by generating a PANI film imprinted with caffeine metabolites on a glass support in order to detect trace concentrations of such compounds in the sweat of a fingerprint. The hybridization of this technology with fingerprint identification technology would provide the capabilities of simultaneous collection of fingerprints and detection of illicit excreted substances within the sweat of the collected fingerprints.

Explosives, Fingerprints, Molecular Imprinting

C35 Cold Case: A Hit-and-Run Driver Identified by an Ad Hoc Pyrolysis GC Device

Luis Chavez, BS, Anfon Muñoz 700, La Serena, CHILE; and Cristian F. Lizama, BS, Arturo Prat 19, Temuco, Araucania, CHILE*

After attending this presentation, attendees will understand that an old problem could be answered by adapting old equipment to new needs. Pyrolysis of solid samples coupled with Gas Chromatography/Mass Spectrometry (GC/MS) chromatography is made normally in specially designed devices which function as an add-on to normal equipment. However, the cost of these devices in developing countries is usually expensive, so in this case a Graphite Furnace Atomic Absorption Spectrophotometer (GFAAS) was utilized as a replacement by being used as the thermal unit.

This presentation will impact the forensic science community by providing forensic laboratories of non-developed countries the chance to use equipment commonly found in analytical laboratories, such as GFAAS, to decompose solid material which is to be analyzed.

This study will suggest to forensic laboratories of non-developed countries the opportunity to use equipment commonly found in analytical laboratories, such as GFAAS, to decompose solid material to be analyzed. In this case, the sample was sealed in a glass chamber under argon atmosphere. The sample was then placed in the graphite furnace device in out-of-service AAS equipment, where pyrolytic decomposition was carried out. A solid phase microextraction unit was placed inside the chamber which, after an adsorption process was inserted at the injection point of a GCMS, was used to identify the thermal decomposition products of the sample. Several previous studies were conducted with different brands of paints of varying colors and dates of application. Samples were collected from the solid matrix and decomposed thermally as described. Chromatographic peaks were compared as retention times and relative heights, showing that the method of processing the sample is useful and reproducible.

The first case solved with this technique was a hit-and-run cold case which occurred in Copiapo in 2010. A 25-year-old young man was hit by a car whose driver left the scene without helping the victim. Several chips of blue car paint were found on the clothes of the man, but at the time no suspect was available. Two years later, a man who was identified by witnesses of the accident as the driver was found with a car of the same make, model, and color as that used in the incident. After a court order, samples of car paint were collected and submitted to the forensic laboratory, where the different layers of paint chips were isolated and analyzed separately. The blue paint was pyrolyzed as described and analyzed by GC/MS, showing sufficient similarity to present to the court the responsibility of the driver.

This case will be presented along with previous studies of paints and coats, showing the feasibility of this procedure.

Pyrolysis, GFAAS, SPME

C36 Hazards of Seat Belt Load Limiters

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After attending this presentation, attendees will understand how seat belt load limiters can present a hazard to occupants in certain crashes.

This presentation will impact the forensic science community by explaining how seat belt load limiters work as well as their benefits and hazards.

History of Seat Belt Load Limiters: A seat belt load limiter controls tension in the seat belt in order to limit the force the seat belt applies to a restrained occupant during a crash. Load limiters can reduce head and upper torso injuries for dummies in frontal crash tests through increased energy management.¹ Load limiting belt systems were first allowed on passenger vehicles in the early 1980s but did not become prevalent until the mid 1990s.² The National Highway Traffic Safety Administration (NHTSA) currently has a Federal Motor Vehicle Safety Standard for seat belts (FMVSS 209). In this standard, there are limits on how much elongation is allowed in the seat belt webbing when a load is applied. In 1981, NHTSA amended FMVSS 209 to exempt seat belts from the belt elongation requirements of the standard if they were installed with an automatic restraint.³ With this amendment, no upper limit on belt elongation was established because NHTSA believed that no manufacturer would design a load limiting seat belt that would elongate appreciably beyond the limits of FMVSS 209.⁴ However, the current design belief is that the amount of elongation, should be larger than the available interior distance between the forward structures and the occupants chest in order to dissipate a larger amount of energy.⁵ This design theory relies heavily on the front airbag to mitigate head injury.⁶ While research conducted by NHTSA on fixed barrier crash tests showed a reduction of injury potential for both head and chest, recent narrow offset crash tests performed by the Insurance Institute for Highway Safety (IIHS) demonstrate an increased injury potential for both head and chest when load limiting seat belts are used.^{7,8} A case study is shown to demonstrate that severe head injuries can result from a load limiter with no upper limit on webbing elongation designed into the system.

Case Study: The restrained driver of a passenger car sustained a debilitating head injury in a narrow offset frontal impact. The Delta V experienced by the vehicle was approximately 38mph. In the crash, the steering column moved inboard. Due to the dynamics of the accident, the driver moved toward the A-pillar, glancing off the deployed airbag, and impacting the structures in the area of the lower A-pillar. The seat belt load limiter allowed the driver's head to make contact with the driver's side A-pillar. This impact caused a severe skull fracture with resulting brain injury. In this event, a limit on the amount of webbing elongation would have prevented contact with the A-pillar structure and prevented the injurious head impact.

Conclusions: In conclusion, seat belt load limiters have been shown to reduce head and upper torso injuries for restrained dummies during frontal impact crash tests, but this same benefit is not realized for humans in high-speed offset frontal crashes. To prevent these serious injuries, a limit on webbing elongation should be utilized with load limiter designs.

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Seat Belt, Load Limiter, Offset Frontal Impact

C37 The Forensic Analysis of an Airbag Deployment Causing Injury in a Non-Collision Event

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The goal of this presentation is to outline the steps used to analyze and evaluate the cause of an unintended airbag deployment that resulted in an injury to the vehicle operator.

This presentation will impact the forensic science community by providing background and illustrating the analytical techniques used to find the cause of an injurious airbag deployment.

The driver and sole occupant of a small, domestic Sports Utility Vehicle (SUV) suffered cartilage and ligament damage to the left wrist, with complications, when the driver-side frontal airbag deployed in a non-collision event. An investigation was undertaken to determine the cause of the unintended airbag deployment that included the review of vehicle maintenance and repair records, inspection of the roadway, inspection of the incident vehicle, and removal of the airbag system components.

The vehicle had been parked parallel to a curb. After entering the vehicle and starting the engine without incident, the driver-side airbag unexpectedly deployed just as the slow moving vehicle was maneuvered a short distance into the roadway. While the vehicle was also equipped with a passenger-side frontal airbag, only the driver-side device activated.

A document review showed the SUV, purchased as a new vehicle, was routinely serviced by two authorized dealers under a factory-specified maintenance schedule. The service records reveal the SUV was involved in two minor traffic collisions. One collision occurred at an odometer reading of approximately 9,000 miles that resulted in minor property damage to the rear quarter panel. A second collision, categorized as a right side T-bone impact, was noted to have occurred at approximately 27,000 miles. Both collisions were not likely to have affected the function of the frontal airbag system; however, the records also show that at about 15,000 miles, the steering wheel was removed and the cruise control switch assembly was replaced under warranty. This switch assembly was damaged by the owner's pet, who regularly occupied the vehicle's interior.

The roadway was inspected where the incident had occurred. No physical evidence related to a curb strike of sufficient magnitude that could be associated with airbag deployment was noted.

The incident vehicle was inspected and thoroughly examined for physical evidence indicative of a frontal impact. None

of the wheel rims showed evidence of a curb strike. There was no evidence of ground contact to the undercarriage and suspension components. Furthermore, no evidence to the reinforcement beam related to a frontal impact of significant magnitude was observed during the tear-down of the front bumper assembly.

The vehicle was scanned for diagnostic trouble codes prior to the removal of the steering wheel and driver-side airbag module, clock-spring, and, Engine Control Unit (ECU). Two fault codes were identified. The first code was related to a low-voltage condition of the battery that had subsequently drained during vehicle storage. A second code identified a system condition related to the deployment of the driver airbag.

A close-up examination of the vehicle components that were removed from the incident vehicle revealed abrasions to the wire's insulation and connector of the clock spring. It was proposed that these abrasions were caused by a misalignment of the steering wheel during the replacement of the cruise control assembly switch. When these two components were realigned, an interaction between the wire insulation and a pin on the back of the steering wheel was observed. Therefore, it was concluded that the reinstallation and misalignment of the steering wheel caused the insulation abrasion that resulted in grounding the wire core to the steering wheel's metal frame.

Frontal Airbag, Clock-Spring, Fault Codes

C38 Injuries Caused by Air Bag-Induced Lower Extremity Flail

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The goal of this presentation is to present case studies that show some consequences of front passenger interaction between the occupant's lower extremities and deploying air bags.

This presentation will impact the forensic science community by presenting case studies that demonstrate a type of airbag injury that has not been previously recognized in the literature.

It is generally accepted that driver- and passenger-side airbags provide a net safety benefit in frontal automobile crashes. However, soon after airbags became more common in the fleet, accident investigators began to report airbag-related injuries, most of which involved minor injury, but also included some serious and fatal injuries.

There are two main categories of injury mechanisms related to airbag deployments: those that result from direct interaction with the airbag during punch out, which are characterized by localized high-force damage patterns; and, those that are caused by interaction with the airbag during the membrane phase, which ordinarily can be characterized as some type of flail. Often such flail injuries are upper extremity orthopedic injuries that result from the forearms being propelled by the deploying airbag into structures, such as the vehicle's roof, door, or dash. When the upper extremity is flung into the occupant's head and face, facial fractures, ocular injuries, and closed head injuries can also be part of the flail injury pattern.

It has been reported that lower extremities are not in the path of the deploying airbag, and it is therefore not surprising that airbag-related leg injuries have not been reported in the medical literature or identified in field accident investigations.¹ Certainly there are circumstances where the lower extremity can be injured interacting with a deploying airbag. Indeed, Huelke reported that a woman seated with her foot on the dash over the module sustained comminuted foot fractures during a frontal crash with

deployment, which would be a punch out-type injury. To add to the understanding of the consequences of interaction between lower extremities and deploying airbags, two cases of lower extremity flail injuries are presented.

Case 1: A full-sized pickup turned left in front of an automobile during an intersection collision. The passenger described sitting with her feet on the dash. The collision was sufficient enough to warrant an airbag deployment. The top-mounted passenger-side airbag deployed and forcefully propelled the occupant's lower extremities into her facial area. The resultant forceful leg-to-head strike caused a brief loss of consciousness, a closed head injury, and, presumably due to bending while the lower extremities were propelled upward and backward, she also sustained a lower back injury.



Figure 1. Vehicle damage from a deer impact.

Case 2: A vehicle vs. deer collision produced deployment of the frontal airbags. Analysis of the vehicle's damage, shown in Figure 1, as well as the impact speed and relative weight of the deer showed that the Delta V was less than 3mph. Clearly, with such a benign impact, the only mechanism of injury would be related to the airbag deployment. Indeed, the driver and rear seat passengers were uninjured. However, the right front passenger sustained a brief loss of consciousness and extensive facial injuries, including a LeForte II fracture, with jaw, nasal and right orbital fractures. Later records showed that she also sustained disruption of her popliteal tendon and the lateral posterior knee capsule of her left knee.



Figure 2. Facial injuries of the passenger in the hospital and two years later.

The passenger was seated with her legs crossed so that her left ankle was on her right knee. In this position, the left leg was evidently in the path of the deploying mid-mounted passenger side airbag. It was concluded that the deploying passenger airbag forcefully propelled the lower leg at a catastrophic speed directly into the passenger's face, causing her extensive head and facial injuries as well as a twisting injury to her left knee. Therefore, this injury pattern could be characterized as a lower extremity flail injury due to airbag deployment.

An examination of two case studies was performed. In these case studies, prior to the subject accident, the right front occupant's lower extremities were located the vicinity of the airbag and/or airbag door. When the airbag deployed, the occupant's lower extremities were forcefully propelled rearward into the occupant's facial areas from interaction with the airbag during the membrane phase. The lower extremity flail-type injuries resulting from interacting with the airbag during the membrane phase resulted in not only lower extremity injuries, but also facial, head, and lower back injuries. In both of these case studies, the injuries sustained would not be expected had there been no interaction with the lower extremities.

Reference:

1. Huelke DF. An Overview of Air Bag Deployments and Related Injuries. Case Studies and a Review of the Literature. SAE Paper 950866, Society of Automotive Engineers, Inc., Warrendale, PA, 1995.

Head and Face Injury, Lower Extremity Flail, Air Bag

C39 Dangers Associated With In-Seat Occupant Sensing Systems

Matthew A. Ivory, BS, 3720 E LaSalle Street, Phoenix, AZ 85040; and Carley C. Ward, PhD, Biodynamics Engineering, Inc, 3720 E La Salle Street, Phoenix, AZ 85040*

After attending this presentation, attendees will understand how flaws in occupant sensing systems can lead to unwanted airbag suppression.

This presentation will impact the forensic science community by explaining how in-seat occupant sensing systems can sometimes call for suppression when an airbag deployment is actually desirable.

History of In-Seat Occupant Sensing Systems: In 2000, the National Highway Traffic Safety Administration (NHTSA) published their final rule for Federal Motor Vehicle Safety Standard

(FMVSS) 208, which is the standard related to occupant crash protection. The purpose of the final rule was to reduce the risk of serious airbag injuries to small women and young children. In the final rule, NHTSA gave manufacturers the option of either developing an airbag capable of a low risk deployment, or provisions for the airbag system to identify the right front seat occupant and suppress deployment of the airbag as needed. In the low risk deployment option, the airbag must deploy in such a manner as to not be injurious to children and small females in a series of tests. Most manufacturers chose the airbag suppression option and developed cushion-based, in-seat sensors to help identify the occupant. With the development of cushion-based, in-seat sensors, it was soon discovered that there was an overlapping zone between the 5th percentile female (152.4cm, 46.3kg) and a 30kg child. In order to remedy this overlapping zone, manufacturers created seat cushion sensors that not only looked at the occupants' weight, but also utilized a pressure map. These systems had difficulty identifying a human versus a dummy occupant due to the pressure map created by the dummy. Because of this difficulty, NHTSA ultimately allowed manufacturers to bypass their suppression system in their crash tests. This created a scenario where a 5th percentile female human could be classified as a child if their buttocks created a pressure map that was misinterpreted by the system as a child, and they would not receive the benefit of an airbag in a crash. Also manufactures are free to develop their own seat-mounted sensing devices for discriminating between a small woman and a child. Because the sensing devices differ according to vehicle model, testing with human surrogates is advised when airbag suppression is suspected.

Case Study: The restrained 19-year-old female, right front passenger of a passenger car sustained a serious brain injury in an impact with a tree. The Delta V experienced by the vehicle was between 35 and 40mph. The right front passenger weighed as much as a 5th percentile female and was the same height, but was nonetheless identified by the in-seat sensing system as a child and her airbag did not deploy. Without the airbag deployment, she traveled forward until her head struck the dashboard. Had the airbag deployed, she would not have sustained permanent and serious brain injury. To test the performance and accuracy of the in-seat sensor, she was placed in exemplar vehicles and the airbag activation light was photographed. Depending on how she sat in the seat, i.e.; orientation of her thighs, the airbag was or was not suppressed.

Conclusions: In conclusion, airbag suppression is helpful in preventing injuries to small children, but in-seat sensing systems can create a situation where a small female is mistaken for a child and is left without the protection offered by airbag deployment.

Airbag Suppression, FMVSS 208, In-Seat Sensing Systems

C40 Determination of Seatback Movement Thresholds Resulting in Hazardous Amounts of Vehicle-Anchored Seat Belt Slack

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After attending this presentation, attendees will understand the phenomenon of how and why vehicle-anchored seat belts are rendered ineffective due to slackening as a result of rearward seat deflection. Techniques for assessing this hazard and determination if belts were worn despite the occurrence of occupant ejection will be demonstrated. Lack of knowledge about this hazard has led to

incorrect assessments of seat belt usage, pre-impact seat position, and driver identity.

This presentation will impact the forensic science community by providing critical information about a significant vehicle hazard which is not adequately addressed by existing motor vehicle safety standards or auto industry practices, and which affects the safety of both front and rear seat occupants.

Dynamic vehicle testing proved in the 1980s that 1" of seat belt slack increases injury by 50% and 4" of slack is essentially the same as being unrestrained in a frontal crash. Collapsing seats typically create 7-14" of vehicle anchored belt slack in rear impacts. Auto manufacturers warn against voluntary seat back or seat track movement when vehicles are in motion because seat movement can lead to a reduction in seat belt effectiveness and increased risk of injury, but most seats and belts cannot prevent these hazards. Crash safety research has proven that conventional belts are not effective at providing occupant restraint in rear impact or rollover crashes where significant seat back deformation and/or seat track movement has occurred. When seat backs reach 40-45 degrees aft of vertical, they can no longer provide effective restraint in a rear impact or rollover, and vehicle occupants begin ramping up the seat back and out of their belts. Seat and belt research by University of California, Los Angeles (UCLA) and the National Highway Traffic Safety Administration (NHTSA), as well as more recent research by Ward, Pozzi, Saczalski, et al., has repeatedly proven this in dynamic testing as well as field investigations. Testing to determine specific amounts of belt slack relative to seat back displacement involved determination of unloaded belt slack generated as a result of maximum available seat back recline, without buckle movement, with various-size adult human surrogates. This proved that larger occupant torso circumference (especially in females) tends to generate the greatest amount of belt slack when the upper torso falls away from the torso belt as a result of seat back movement. The resultant torso belt slack is then free to pass to the lap portion of the belt, especially if belts are equipped with pass-through latch plates. Rearward movement of buckles and hardware causes additional belt slack. Belt slack and rearward hardware movement as a result of seat deformation are contrary to the original intent of FMVSS 209, Section 4.1, which required that the lap belt remain on the pelvis at all times during collision or rollover. This is a well-recognized essential foundation for any effective belt restraint system. It is not good safety practice to knowingly design and install weak seats which will predictably create dangerous seat belt slack under foreseeable impact loading, contrary to warnings to vehicle occupants.

The purpose of this study was to scientifically determine whether lesser amounts of seat back movement can create significant slack in conventional belts which would significantly increase the likelihood of occupant injury and/or ejection. Front bucket seat backs in various vehicles were initially positioned at the full upright adjustment position, approximately 23 degrees aft of vertical and the seat track in a position to allow comfortable surrogate hand and foot contact with the vehicle controls. The surrogate entered the seat and applied the seat belt properly with available retractor tension and no slack. The seat belt was then clamped at the B pillar "D" ring to simulate retractor lock-up and prevent any further movement of the webbing. The seat back was reclined until the surrogate could consistently create sufficient slack to slide rearward up the seat back and out of the lap belt. The seat back deflection, belt slack, and occupant movement potential relative to the lap belt and vehicle interior were documented. Testing showed that static seat back deflection approximately mid-way between upright and full reclined would generally result in sufficient belt slack to allow complete ejection of the belted occupant out of the restraint system. A typical front seat back reclines from approximately 23-25 degrees to 45 degrees or more aft of vertical, with an approximate movement range of 20-22 degrees or more. The "threshold" of

10-12 degrees of static seat back deflection appears sufficient to disable many conventional vehicle-anchored belts.

Rear Impact, Seat Belt Slack, Seat back Movement

C41 Failure Mode of an Aircraft Restraint

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After attending this presentation, attendees will review a failure mode in a general aviation shoulder harness retractor. Aircraft accident investigators will learn how to check for this failure mode and the cause of the failure.

This presentation will impact the forensic science community by demonstrating different injury patterns of similarly restrained occupants in an aircraft crash may be the result of many factors, e.g., position, body characteristics, age, intrusion, and, of course, restraint failure. This presentation focuses on a restraint failure mode and how to detect and document it.

The right front passenger was severely injured when the four-seat, high-wing aircraft crash landed due to a loss of power from a fatigue failure of the No. 4 cylinder. The plane broke apart as a result of the crash.

The single-engine plane was flying at about 3,500 feet when the occupants heard a loud bang and felt the plane shake. The pilot guided the plane toward a helicopter landing pad near a beach in southern California. The plane touched down on flat ground, then collided with the upslope side of a ditch and broke into pieces.

The restrained front passenger was about 196cm in stature and weighed approximately 117kg. His seat was pushed all the way back. This left no foot room for the restrained rear passenger who had to sit sideways in the rear seat with his feet on the left side and his back against the right side. The pilot was slightly smaller, 183cm, and weighed about 92kg.

Both front seats were equipped with three-point restraints in an unusual configuration. The lap belt buckles with end-push release were mounted on the outboard side of the seats with 36° upward travel from horizontal. The Automatic Locking Retractors (ALR) were on the inboard side. A double Emergency Locking Retractor, Web sensing (ELR-W) was mounted to a frame member in the roof. The right retractor was on the left side and the left retractor was on the right side with the shoulder webbing crossing each other near the roof. The other ends of the shoulder belt webbing were attached to fixed latch plates.

ELR-W retractor locks were activated by a rapid extraction of the webbing off of a storage reel in the range of 0.75g to 1.5g. To check the retractor locking activation operation, the webbing was quickly pulled out. The pilot's shoulder belt reel locked and the front passenger's did not.

Injuries to the passenger were consistent with a lack of upper body restraint. They included contact with the roof liner and windshield header. Witnesses noted that the front passenger was found partially outside the aircraft with the shoulder belt extended and around his neck.

To understand the nature of the failure, the retractor inertial locking mechanism was examined. This mechanism consisted of a centrally pivoted plastic arm attached to the rotating reel. The arm was held in place by a calibrated spring that had an engaging tooth that swung out against a plastic ring with internal fixed teeth. When rotational forces overcame the spring tension, the arm extended into a plastic ring that had two levers that rotated slightly when the arm locked and engaged a lock bar. This moved the lock bar into two sets of metal teeth attached to the webbing storage reel, stopping it.

When the sensor mechanism cover was removed, it was noted that the arm on the plastic ring had broken off so that it could not move the lock bar into locking position. The storage reel teeth cut a groove into one side of the lock bar and tooth tips were sheared off as the lock bar tried to engage the reel teeth. This phenomenon is also known as skip lock. This appeared to be a manufacturing error instead of a design error.

A detailed examination showed the retractor's failed components, and witness marks were found within the subject retractor.

Investigators must look at not only the equipment and its function but occupant kinematics, injury patterns, and witness statements to fully understand the event.

Seat Belt, Retractor, Aircraft

C42 Urban Design and Crime: The Problem in a Megalopolis

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The goal of this presentation is to explain how reducing crime and the fear of crime may be possible by proper environmental design.

This presentation will impact the forensic science community by increasing awareness of the fact that urban design and proper planning are important elements in developing crime prevention strategies.

This presentation will introduce a new criminological theory that supports the idea of a correlation between the organization of urban space and crime. While it may be possible to achieve crime prevention through urban space design, it has to be included in the strategies of city planning. Such planning can reduce not only the opportunity to commit crime, but also lessen the motivation to offend, achieving more attractive public physical environments. An awareness of rising activity is essential and requires primarily the research of current crime data, with the ultimate goal of implementation based on that aspect. National guidelines, implemented by local governments, should be developed based on the results obtained. Istanbul is a megapolis with a population approaching 20 million, and is a well-known city centuries old that has served as the capital of empires and continues to be the most important city of the Turkish Republic. It is very difficult to re-plan a historic city, but in reality it is dynamic and changes constantly. Therefore, urban planning with the goal of reducing crime rate is possible and essential.

Based on the fact that crime rates are very much culturally dependent and environmentally affected, the goal of this study is to evaluate two types of crime in an older, larger district of the city of Istanbul called Zeytinburnu (293,228 residents as of 2011). Data was collected and mapped on burglary and prostitution cases from the law enforcement database for a one-year period. The results showed that the crime rates vary significantly between different suburbs. Following this information, an attempt was made to determine the characteristics of urbanization, the building statute, and to evaluate them in terms of building design and lighting in the

above-mentioned area. There are primarily two types of residential settlements in the district in question: (1) the classical "old type" neighborhoods; and, (2) the "housing complexes" which are guarded and very well protected by numerous types of technical protection units (cameras, videos, etc). An interesting result obtained was not being able to deduce an outcome and generalize it for all the crime types. As expected, in this study, the very well-protected "housing complexes" certainly had fewer burglary cases. However, the over protected and technically observable residential areas were the most preferable areas for prostitution, because the surveillance equipment served as information-delivering systems for prostitution protection.

It is true that fear of crime affects people's quality of life. Certainly, the role of law enforcement is important, but equally important are the strategies for crime prevention. Within the various practical applications for crime prevention, improving the urban environment will seriously reduce the criminal opportunity even in large and overpopulated cities; however, it should always be kept in mind that crime prevention is a very complex issue.

Crime Prevention, Urban Design, Istanbul

C43 How One Small Forensic Business Has Contributed to Forensic Science Education and Mentorship

James Millette, PhD, MVA Scientific Consultants, 3300 Breckinridge Boulevard, Ste 400, Duluth, GA 30096*

After attending this presentation, attendees will understand how a small forensic business can provide mentoring and education for aspiring scientists.

This presentation will impact the forensic science community by inspiring other companies to become more involved with forensic science education and mentoring efforts.

MVA Scientific Consultants is an independent testing and consulting laboratory with a staff of 15 part-time and full-time employees. In the past 23 years of existence, it has had over 40 paid interns and a number of unpaid interns who received college credit for their time at work. Because the education level of the interns has varied from high school to graduate student, the mentor with each intern has balanced the needs of a particular project with the intern's capabilities. Beyond washing laboratory glassware and cleaning up the warehouse, the interns have assisted in a wide variety of projects. Some interns have assisted with pharmaceutical contamination investigations, while others have concentrated on projects involving the World Trade Center Disaster, the aftermath of Hurricane Katrina, the Tennessee Valley Authority fly ash spill, dust in the air from Air Force tests of burning air planes, dust from the Alamo Dome Super Fund site in Texas, unknown particles from an intensive care newborn facility in Tennessee as well as moon dust, comet dust, and dust collected floating around in the space shuttle. Interns have received training and become operators of light and electron microscopes, as well as users of the infrared and Raman instruments. One intern spent the summer trying to figure out ways people might cheat on scratch-off lottery tickets. One intern won a scientific microscope photography contest with a submission of a 3D cenosphere fly ash particle that was taken with a scanning electron microscope during an environmental forensic project. Another intern won the annual company holiday card contest with an image of a talc particle that was colorized to look like a Christmas stocking. A collaboration between MVA Scientific Consultants and Gwinnett Technical College's Bio-technology program has provided a number of students with the opportunity for on-the-job training and commercial business hours they needed to complete their technical degree. Many of the interns have gone

on for more education in a scientific area and some have eventually become employees of MVA. MVA Scientific Consultants has also contributed to forensic science education by hosting the Georgia Microscopical Society Young People's course on Microscopy for over 20 years. Staff members of MVA join other members of the Atlanta area microscopy community in teaching this free course for middle-school students and teachers on seven Saturday mornings in the winter. In addition to basic instruction on how to use a microscope, the course includes sessions on criminal forensic and environmental forensic microscopy, as well as geology, biology, and photography through the microscope.

Mentors, Forensic Education, Internships

C44 Thermographic Applications to Pharmaceutical Industry Monitoring and Detection of Counterfeit Drugs

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The goal of this presentation is to propose a new and innovative methodology to be used for monitoring and detection of counterfeit drugs, providing attendees with an increased knowledge of monitoring and detection techniques.

This presentation will impact the forensic science community by sharing a proposed methodology based on the use of thermal infrared imagery. This will be acquired with the last-generation IR thermal scanners. A library of a number of reference drugs signatures, to be compared with those coming from the counterfeit ones, will be suggested.

According to World Health Organization (WHO), counterfeit drugs are products with the correct ingredients or with the wrong ingredients, without active ingredients, with insufficient active ingredients, or with fake packaging. Public health risks include ineffective medicines, intoxication due to harmful ingredients, no quality assurance, no regulatory oversight, adverse reactions not monitored, product recall not possible, erosion of public confidence in medical care and health systems, and waste of money.

Counterfeit medicines pose an ever-increasing threat to public health. Precise tracking of illegal counterfeit prescription drug activity is next to impossible. Low-quality counterfeit medication may cause several dangerous health consequences, including side effects or allergic reactions, in addition to their obvious lack of efficacy due to having less or none of their active ingredients.

Currently, there are no ultimate solutions to the problems associated with counterfeit drugs. The goal of this work is to show the application of a thermography Infrared (IR) system for determining the authenticity of a pharmaceutical product. That application was first proposed by two inventors in Israel and patented. They proposed a method and system for determining the authenticity of a pharmaceutical product using thermography-based equipment.

It is possible to determine counterfeiting even without removing the product from its cover, package, or container. The system to test the authenticity of a pharmaceutical product is composed of a thermography IR apparatus for acquiring, in a wavelength or a wavelength range selected from the Mid Wave IR (MWIR) to very Long Wave IR (VLWIR) range, one or more thermographic IR images of a certain product. A database of predetermined signatures of reference drugs, and a display unit for displaying at least one or more IR images and a signature of a predetermined reference drug or a result of comparison between one or more IR images and a signature of a predetermined reference drug are required.

A study was conducted on the existing technical scientific-

related literature, with the goal of focusing on the new applications of the IR thermography technology already vastly adopted in industrial and, especially, chemical facilities for production and maintenance activities. Evidence of the topic in question has been found in numerous similar problems at the international level. The result of this extensive research of applications has led to finding a promising new investigation methodology for the detection of the counterfeit drugs on the market, on which this study was based.

At the international level, results suggested a database of all known sources of counterfeit drugs, keeping track of underground counterfeiting operations, and divulging all information to international anti-crime agencies. Thermography allows detection of the counterfeit phenomenon, whether a suspicious sample is original or not. The methodology requires the use of a specialized laboratory to rapidly screen the increasing suspicious samples, detect counterfeit drugs, and allow the seizure of these products. Legal prosecutions perform more in-depth analyses on some carefully selected samples, evaluate the real risks with the further evolution of this phenomenon, and also update the data for informational campaigns.

The use of IR technology (already developed for a chemical industrial plant) on a major number of products will make the proposed methodology even more reliable and broadly applied.

Drugs, Counterfeiting, Thermography

C45 Output Characterization of Handheld Lasers Used in Criminal Aircraft Illumination

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After attending this session, attendees will understand the basics of laser metrology, laser safety, and adverse visual interference effects that can be generated as they pertain to the evaluation of handheld lasers used in criminal aircraft illumination cases.

This presentation will impact the forensic science community by demonstrating a method for easy, rapid, and accurate characterization of handheld lasers used in criminal aircraft illumination cases, and to provide quantitative data to be used in the judicial process.

In providing quantitative data on the devices used, personnel involved in the prosecution of cases will be better able to educate those involved in the judicial process of the potential hazard that these devices present. Illumination from a hand-held laser can, at short range, induce permanent retinal damage. At long range, as in the cases of aircraft illumination from the ground, disruptive visual interference effects can occur. Effects such as flash blindness, after-image, and glare are possible from even relatively low-powered devices. Threshold irradiance values have been established by the Federal Aviation Administration (FAA) for varying degrees of visual interference effects.¹ This measurement system will allow for calculation of this irradiance from the devices in question.

The measurement system discussed will evaluate the devices in two ways. The first is to establish the actual optical power emitted from the device.² Recent studies have demonstrated that the majority of commercially available "laser pointers" are emitting laser radiation in excess of the federal limit, and in many cases, well in excess of the claimed output power shown on the labeling on the devices themselves.^{3,4} This measurement system will enable easy and accurate evaluation of the output power. Since the degree of

the hazard the devices pose is proportional to the optical power emitted, providing data on the actual power emitted (not simply what is claimed on the label) is essential.

The second aspect will provide a measurement of the divergence of the visible beam emitted from the laser. Since this system is to provide quantitative data for the prosecution of laser-aircraft illumination cases, it is important to provide accurate data for calculation of the irradiance at range. This divergence value, used in conjunction with the output power measured and the flight conditions recorded at the time of the incident provides the information needed to calculate the exposure conditions at the time of the incident.^{5,6} The irradiance calculated in this process allows for a quantitative presentation of the potential short-range hazard and long-range visual interference effects these devices have when illuminating an aircraft.

This system is presented as a measurement solution that could be implemented easily within the law enforcement organizations tasked with prosecution of these cases. The design that will be discussed will emphasize ease of use, accurate results, and moderate cost to establish a measurement system. The end result will enable a better education on the potential hazards that hand-held lasers can create, and to promote that knowledge to those involved in the judicial process of criminal aircraft illumination cases.

References:

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2. Meas. Sci. Technol. 24 045202 (2013)
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4. J. Laser Appl. 25, 032007 (2013)
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6. ANSI Z136.6-2005 American National Standard for the Safe Use of Lasers Outdoors

Laser Pointer, Aircraft, Measurement

C46 Human Factors Evaluation of Entry Step Switch in a Recreational Vehicle

Kenneth Nemire, 1840 41st Avenue, #102-242, Capitola, CA 95010-2527*

After attending this presentation, attendees will gain an understanding of human-machine interface issues related to the arrangement and coding of controls, as well as insight into human perceptual and cognitive capabilities and limitations that often interact with engineering issues.

This presentation will impact the forensic science community by demonstrating the practical application of human factors research in the evaluation of a litigated personal injury case.

The findings of a human factors evaluation of an incident in which a woman was seriously injured when she stepped out of her Recreational Vehicle (RV) at night, not realizing the power entry step was locked in the retracted position will be presented.

The evaluation included analyses of the plaintiff's operation of the entry step control, the design and operational logic of the entry step control, the arrangement of the entry step control, and the adequacy of warnings adjacent to the entry step control.

Evaluation of the plaintiff's behavior and the operational logic of the power entry step control revealed the following:

1. Wvnd she fell to the ground.

A different operational logic design could have prevented this incident. A later version of this RV provided an operational logic that made it impossible to lock the power entry steps in the retracted position.

Evaluation of the entry step control revealed that it was a rocker switch located near the door and adjacent to three other

rocker switches, one of which controlled the porch light. Analyses of previous human factors engineering research and human-machine interface design guidelines, which will be discussed during the presentation, revealed that the arrangement of the entry step switch, as well as the similar coding for the four switches, made accidental operation of the entry step switch a foreseeable event.

Three of the switches controlled power to three different lights, and the fourth controlled power to the entry steps. Pressing the entry step switch ON did not extend the steps as the ON position of a light switch turned on a light; however, the plaintiff thought of the entry step switch in the same way as one would think of a light switch. The different operation of the entry step switch required that it be arranged and coded differently from the light switches to facilitate user understanding; however, it was not.

Given the confusing nature of the different switch operations, a warning would have been useful. However, the existing label warned about accidentally retracting the steps by turning on the engine ignition switch while the RV was parked, but failed to mention other ways to cause the entry steps to retract, such as switching the entry step power OFF. Other issues with the failure to provide format, content, and location of an effective warning will be provided during the presentation.

This injury incident was primarily caused by: (1) a poorly designed operational logic; (2) the similar coding and close proximity of controls for the entry step and porch light; and, (3) the failure to warn.

Human Factors, Human-Machine Interface, Warnings



General



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D1 Evaluation of Alumni Contact in Forensic Science Programs

Catherine G. Rushton, MS, 1401 Forensic Science Drive, Huntington, WV 25701*

After attending this presentation, attendees will evaluate ways to maintain communication with alumni. Increased communication with alumni will benefit faculty, students, alumni, and laboratory personnel.

This presentation will impact the forensic science community by seeking to bridge the gap between academia and forensic laboratories. Alumni of forensic science university programs gain employment in forensic laboratories which allows them the opportunity to assess the strengths and weaknesses of forensic science curriculum in light of knowledge and skills required by their job.

Current students become alumni when they graduate. They share similar experiences but at different points in time. Because alumni have traveled the same path that current students are walking, alumni are a resource to assist current students and faculty. This resource is only useful if communication between alumni and the program exists.

In order to make better use of this valuable resource and better evaluate success of the graduates in their jobs, the forensic science programs try to remain in communication with alumni. Graduate and employer satisfaction surveys allow faculty an opportunity to evaluate whether or not their classes are preparing the students for their jobs based on the answers from the graduates who have been employed for at least a year and their immediate supervisors. Faculty are also able to call upon the expertise of alumni, employed in the forensic science field, to offer their advice, guest lecture, attend seminar series, or judge research posters. Alumni benefit from remaining in contact with the program with opportunities for collaboration on research projects. They also benefit from research conducted by students in their laboratories when they host an intern. Laboratory personnel benefit from increased communication by being able to advertise services and continuing education opportunities offered by the laboratory.

Forensic science programs at several universities struggle to maintain meaningful contact with their alumni. The most common methods many programs use to maintain contact with alumni are surveys, Facebook®, newsletters, personal communications, alumni receptions at national meetings, and emails. Prior to graduation, the students are informed of ways to stay in contact with their programs when they move from place to place, job to job, change email addresses, or change cell phone numbers. However, during all these life changes, alumni forget to inform the program of the changes. This hinders communication with the alumni. Since continued contact with the alumni is a problem, finding the answers for why alumni do not maintain continued contact can be tricky. Those that are most likely to return an emailed survey are already in continued contact with the program. For those who would likely not return a survey, alternative methods of contact need to be identified.

Literature on alumni relations gives insight into the

general principles involved in maintaining a high level of contact with graduates. Improvements identified in the literature include: including alumni in program activities and events with students; including alumni in advisory positions with student organizations; creating an electronic newsletter; and creating online continuing education courses. These principles could be adapted for use by the forensic science program.

Successful strategies for improving communication would benefit forensic science programs. Ultimately turning the alumni into stakeholders for maintaining communication with the program is a worthy goal. The improvements proposed for increasing communication with the alumni should be implemented in phases. Prior to implementation, a representative sample of the alumni should be surveyed to ensure the improvements will meet their needs as well as the program's needs.

Alumni, Higher Education, Communication

D2 Fingerprint Enhancement on Used Prophylactics

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The goal of this presentation is to demonstrate that because prophylactics can be used in several clandestine manners, when discovered, efforts should be made to process them for latent fingerprints to possibly help identify an offender.

This presentation will impact the forensic science community by strongly encouraging crime scene technicians and criminalists to properly collect, retain in evidence, and process used prophylactics. The most conventional methods of evidence packaging and fingerprint enhancement were relatively ineffective and, therefore, more advanced methods of fingerprint development are encouraged.

The enhancement of fingerprints on used condoms is significant because condoms are used in two clandestine manners: (1) condoms are used to transport illicit drugs and other contraband by being ingested or concealed within various body cavities; and, (2) some sexual predators sometimes use condoms during the perpetration of their sexual assaults. Crime scene technicians discovering used prophylactics may not find it significant at the time to collect, retain as evidence, or process used condoms for latent fingerprints.

This research focused on using the four most conventional methods of latent print development to determine if prints could be developed on used prophylactics. The first of the four methods involved cyanoacrylate ester fuming, followed by cyano-blue dye stain, and then examination with an alternative light source. The second method was cyanoacrylate ester fuming, followed by magnetic powder brushing. The third method used Small Particle Reagent (SPR). The final method involved cyanoacrylate ester fuming, followed by staining with Sudan Black dye. All four methods were studied in an attempt to develop latent prints on six different styles of three major brands of condoms. The scope of this research excluded the ninhydrin process since numerous

studies have used this as the method for latent print development on latex gloves. The results from this study indicated the four most conventional methods used were highly effective for quality latent development on any of the samples, even though minimal ridge definition was developed.

Six styles of three major brands of condoms were used in this study. The condoms were divided into two groups and a latent print was placed on each condom in each group. A sebaceous standard was used to keep prints consistent. Regarding Group 1, a print was placed 25 days prior to processing and placed into evidence. This was done to assess whether delayed processing detracted from the enhancement process. Each condom was then individually packaged in a paper envelope during storage and labeled, pending the enhancement process. The 25-day retention period was selected as the arbitrary length of time that evidence might be in storage prior to undergoing an enhancement process. With the second group (Group 2), a print was placed 24 hours prior to processing. Twenty-four hours was the arbitrary time established for a condom that might be recovered at a crime scene or from a suspect and require immediate laboratory analysis for latent prints. Again, the samples were placed in individual paper envelopes. Researchers used the right index finger coated with a sebaceous film as a standard on the Group 1 25-day samples. When the second sample, Group 2, was prepared, the same researcher used his right thumb coated with a sebaceous film as a standard.

One type of each style of condom from each group was processed with the four different development methods. The first analysis method (A) involved cyanoacrylate ester fuming followed by cyano-blue dye stain and then examination with a alternative light source used in the visible wavelength range from 380nm–470nm. The second method (B) was cyanoacrylate ester fuming followed by magnetic powder brushing. The third method (C) used SPR. The final method (D) used cyanoacrylate ester fuming followed by staining with Sudan Black dye. The results of these methods are seen in Table 1 and Table 2 below.

There was no significant difference in latent print enhancement between the condoms tested in the 25-day sample and the 24-hour sample. Even though the enhancement methods conducted in this study were basically ineffective, it is strongly encouraged that prophylactics found at crime scenes and/or being used for clandestine smuggling should still be processed for possible latent fingerprints. Air drying individual prophylactics prior to packaging as evidence is strongly recommended to prevent destruction of ridge definition caused by a condom adhering to the packaging material and to itself.

Additional research conducted on used prophylactics is strongly recommended, using such methods as Vacuum Metal Detection (VMD) and other contemporary chemical processes.

Clandestine Smuggling, Sexual Assaults, Latent Fingerprint Enhancement

D3 Pulmonary Cement Embolism After Kyphoplast: A Case Report

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After attending this presentation, attendees will be able to collect useful data relating to the possibility of polymethylmethacrylate (PMMA) cement embolism following kyphoplasty (KP), often unrecognized and confused with embolism

by Central Venous Catheter (CVC). This presentation will enrich the scientific community since there are very few reports about this topic.

This presentation will impact the forensic science community by showing how, as in the case of a claim for damage compensation relating to embolism after KP in a CVC bearer, it is important to carry out all necessary investigations to ascertain and clarify the nature of the embolus.

Background: Kyphoplasty (KP) and vertebroplasty (VP) are very common procedures used to treat vertebral fractures. Pulmonary Embolism (PE) is one of the most feared complications of these procedures, with rates ranging from 3.5% to 23%. Local cement leakage and pulmonary cement embolism are quite rare complications, more frequent after VP than KP. Symptoms related to PE were noted in only 0.4% to 0.9% of cases. In fact, in the vast majority of patients, PE after KP is often discovered incidentally by post-operative chest X-rays or Computed Tomography (CT) carried out for other purposes. The literature instead reports cases of PE by CVC occurring more frequently. A postoperative chest radiograph is routinely obtained to confirm the position of the CVC and to rule out complications due to a fractured CVC with embolism of the distal fragment which may lead to catastrophic complications. The diagnosis of the nature of the embolism, as in the case reported by this study, is important not only for clinical purposes, but also for the related legal issues.

Case Report: A 64-year-old man suffering from a lumbar fracture due to injury underwent neurosurgery (KP) with the use of PMMA osseofix. On the seventh postoperative day, the man showed dyspnea. A CT of the chest was performed and revealed the presence of a pulmonary embolism from a foreign body. This finding led to the belief that it was a fragment of the CVC tip as this is a potential and quite common complication. The foreign body was removed by open thoracic surgery following the failure using the percutaneous approach. During surgery, in the apical medium portion of the right pulmonary, a pluri-fragmented whitish colored element was found. After being discharged from the hospital, the subject made a claim for damages against the hospital. The hospital then ordered an investigation to understand the nature of the surgical specimen. This material was examined by spectrometry to understand the nature of embolism (either CVC fragments or PMMA cement). The examination identified parts of the PMMA used for the KP.

Discussion and Conclusion: The possibility of cement embolism after VP and KP should be considered in all cases of dyspnea following neurosurgery. The presence of PMMA in pulmonary blood vessels is under-reported in literature, but it is quite common in clinical practice and, if not diagnosed early, may lead to serious consequences for the patient. There are reported cases in literature in which the cement was lodged in the pulmonary vases for long periods of time without causing damage. As in this case, when it's necessary to understand the true nature of the embolism, the adoption of a chemical investigation provides useful data for a sure differential diagnosis. The diagnosis is useful not only for clinical but also for medicolegal purposes. In fact, in the case of CVC-fragment PE, the patient could make a claim for damages to the manufacturer for a defective medical device and the manufacturer has to compensate for the damage. In the case of PMMA embolism as in the case reported, once it is demonstrated that it was a complication, the damage does not require compensation.

Kyphoplasty, Pulmonary Embolism, PMMA

D4 DNA Typing for Identification of Korean War Victims

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After attending this presentation, attendees will learn about DNA analysis of Korean War victims.

This presentation will impact the forensic science community by providing optimized methods for identifying old skeletal remains.

To identify the remains of service members missing in the Korean War, a population-based and DNA-focused identification system was implemented in 2000 by the Ministry of National Defense Agency for Killed in Action Recovery and Identification (MAKRI) in Korea. Since 2007, DNA analysis of Korean War victims has been performed by the Ministry of Nation Defense Criminal Investigation Command (MND CIC).

Skeletal remains are the only biological material available for DNA typing in the identification of Korean War victims. Therefore, the ability to recover DNA from bones exposed to a variety of environmental conditions has become a valuable tool for identification of Korean War victims. The potential to recover DNA profiles from bone samples has increased since efficient DNA extraction methods and amplification kits are now available. However, the analysis of highly degraded old bone samples is a great challenge for identification of Korean War victims, due to the low level of endogenous DNA, environmental DNA damage, and the potential presence of inhibitors that co-extracted with DNA. To solve multiple problems encountered during DNA analysis of skeletal remains from Korean War victims, several methods have been optimized, including the DNA extraction method, the purification method, and multiplex assay for mtDNA sequencing.

DNA profiles obtained from skeletal samples of Korean War victims are put into a database for random matching and kinship analysis to confirm the relationship between missing casualties and their alleged relatives. Special software was created to compare DNA profiles in the databases of missing casualties and their relatives to detect matches. It has been reported that the problem of the possibility of erroneous matches between unrelated people became significant, especially with the growth of the number of genotypes in the A-STR database. Therefore, increased numbers of markers are required in order to solve this problem. A population study was performed using 23 A-STR markers including additional European standard markers in the Korean population. A combination of 23 A-STR loci provides a dramatically increased likelihood ratio and a valuable tool for the identification of Korean War victims.

This research has been focused on solving problems encountered during the identification of Korean War victims and promoting this national project for the last several years. However, about 130,000 Korean soldiers still remain missing from the Korean War. In order to ensure mission success and the return of unaccounted-for casualties, there will be continued support for MAKRI's personnel accounting efforts.

Korean War Victims, DNA Typing, Skeletal Remains

D5 Development of an Improved Extraction and Analysis Technique for Human Scent Evidence Analysis

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After attending this presentation, attendees will understand principles on how human scent can be used as evidence and how an improved extraction and analysis method lowers the detection limit of human scent residues.

This presentation will impact the forensic science community by demonstrating that human scent residues can be collected, analyzed, and differentiated at lower than-ever-before levels, allowing for the detection of smaller human scent traces.

The human body produces odors made up of a variety of Volatile Organic Compounds (VOCs), which are a class of chemical compounds that have high vapor pressures at room temperature. The combination of these VOCs in different abundances produces a scent, referred to as human scent, which is different for each individual. Human scent is defined as the most abundant VOCs that are identified in the headspace of a collected scent sample. The VOCs that constitute human odor have been shown to be qualitatively similar among individuals. However, the quantitative abundances of individual VOCs and their relative ratios make them characteristic to the individual from whom they are derived. The study of human scent is of substantial interest to the medical and forensic science communities. In the medical community, the VOCs released by the human body can be used as diagnostic tools for diseases. Previous research has identified VOCs that can serve as biomarkers for different diseases. In the forensic science community, human scent evidence is used as an investigative tool that allows for the use of human scent-detection canines in law enforcement.

The current standard for the analysis of VOCs from human scent samples is Headspace Solid Phase Microextraction (HS-SPME), followed by Gas Chromatography/Mass Spectrometry (GC/MS). HS-SPME is a fast, simple, solvent-free extraction method and is suitable for the extraction of VOCs as it reduces interferences due to the sample matrix. However, SPME is a non-exhaustive, highly selective technique and some compounds present in the sample might not be extracted. Also, the aging of the fiber from repeated use results in low reproducibility and this would be disadvantageous when quantitative measurements are required.

Therefore, the current method was developed using dynamic headspace concentration for the recovery of the VOCs from human scent samples. Similar to SPME, dynamic headspace extraction is also fast, simple, and solvent-free; however, this manner of sampling is also exhaustive, providing a more accurate representation of the sample's chemical composition. Also, as degradation of the SPME fiber is not an issue, it can exhibit higher reproducibility and better accuracy when quantitative measurements are obtained. For this work, a standard mixture of compounds that have been previously observed in human scent samples was prepared. This mixture was spiked on cotton gauze, extracted using dynamic headspace extraction, and analyzed using GC/MS. Various extraction parameters were assessed to determine the optimum conditions to ensure the best recovery of the compounds in the standard mixture. Following this, hand odor samples were collected on cotton gauze. The VOCs collected were extracted by dynamic headspace, using the optimized parameters, and analyzed by GC/MS. These results were compared to results obtained using SPME-GC/MS. The dynamic headspace method

was able to extract a wider range of compounds and a lower limit of detection was achieved.

Human Scent, Dynamic Headspace, Volatile Organic Compounds

D6 Is There a Correlation Between Footstep Length, Lower Extremities, and Stature?

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After attending the presentation, attendees will be able to recognize that a possible correlation of stature and footstep length is expected and, consequently, the stature may be estimated from footstep length.

This presentation will impact the forensic science community by recognizing the possibility of correlation observed between footstep length, stature, and thigh lengths, and that the length of the lower limb may not be a major factor in determining the footstep length.

Forensic podiatry is the application of sound and researched podiatric knowledge and experience in forensic investigations. Footprints are important sources of evidence found at various crime scenes. Careful examination of these footprints can provide an abundance of information pertaining to step length, position of each footprint, the shape and size of the footprints and toes, ball lines, instep region, inner and outer margins, injuries, pits, deformities, or accidental damages. Detailed examination of footprints at the crime scene is a vital practice in criminal investigations. Careful scientific analysis of these prints may yield information about the biological profile of the suspect that can help in identifying the suspect and confirm his/her involvement in the crime. A possible correlation of stature and footstep length is expected and, consequently, the stature may be estimated from footstep length.

The present research was conducted on 142 healthy young adults to study the correlation of footstep length, with length of lower extremities and stature in individuals walking at a normal pace. The participants were asked to step on an ink-soaked stamp pad and then walk on a paper roll that was rolled open on an even surface next to the stamp pad. The participants were asked to walk four to five steps in his/her normal stride (pace) on the paper sheet and foot impressions were obtained. Straight distance between the most forward point of the great toe of one footprint and the most backward point on the heel of the next footprint was measured as a footstep length. The first and the last footsteps were not considered to ensure error-free results. The mean of the second and third footsteps was taken as the average footstep length. Stature was measured using a stadiometer. Lower limb length was calculated by subtracting the sitting height from the stature of the participant. The participants were asked to sit erect on a flat wooden stool kept against the stadiometer and the distance between the vertex and the seated buttocks at the level of the stool were measured by using the stadiometer as sitting height. Measurement of the lower limb length was thus an indirect measurement. The thigh length was measured as the distance between the back of the buttock and the proximal edge of the patella. The thigh length measurement used in the present investigation was therefore inclusive of the length of the hips. The leg length was measured as the distance between the anterior surface of the thigh and the floor and included the vertical height of the foot. All these measurement were taken on participants in a sitting posture with hip and knee flexed at right angles using standard techniques and instruments. The study observed that though significant sex differences exist in the stature and length of lower extremities, the step length measurements

were similar among males and females. A statistically significant correlation was observed between footstep length, stature, and thigh lengths in females. However, no such correlation existed among males. The present investigation tests the validity of a proposed hypothesis regarding the relationship between footstep length, length of lower extremities, and stature and suggests that the length of the lower limb may not be a major factor in determining the footstep length of a person walking at a normal pace.

Forensic Podiatry, Footstep Length, Stature

D7 An Unusual Electrocution Death From a Homemade Medical Device

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The goal of this presentation is to discuss an unusual mode of electrocution from a homemade apparatus. After review of this material, the attendees will also understand the importance of obtaining the decedent's medical history and psychosocial information in manner of death determinations.

This presentation will impact the forensic science community by highlighting how pertinent questions of family/friends and acquisition of psychosocial history is important in delineating the circumstances of death.

A man was found dead in his garage with an apparent homemade apparatus attached to his torso. The apparatus consisted of two aluminum pie tins, each connected to electrical wiring that led into an electrical plug. The electrical plug had been inserted into a 120-volt outlet. One pie tin had been placed on the chest, and the other tin had been placed on the back roughly in line with the heart. Examination of the body revealed burns on the chest and back corresponding to the placement of the pie tins. Investigation of the body and the attached device indicated electrocution as the cause of death although the manner of death was uncertain. The man's spouse was questioned regarding his possible intended use for this apparatus. She informed investigators that her husband had been very distrustful of physicians and tended to self-diagnose and self-treat. His lack of trust in established health care was demonstrated by his tendency to research drug information on the internet in order to determine his own medication doses. His medical history had been significant for atrial fibrillation; however, he had avoided following through with medical treatment for this condition. Examination of the man's personal computer uncovered engine searches for purchases of a defibrillator. With this information, it became clear that the pie tins were serving as makeshift cardioversion paddles that, when connected to an electrical source, were functioning as a cardioversion machine. Presumably, this man had intended to substitute his own treatment for his atrial fibrillation rather than submit to a physician's treatment.

Standard cardioversion usually involves low-energy electrical shocks sent through paddles or adhesive pads applied to the chest placed at the right subclavicular area and at the location of the apex of the heart. The electrical shock lasts for a short period and occurs at a specific point in the cardiac cycle to avoid the r wave in the QRS complex. Unintentional discharge on the r wave may result in ventricular fibrillation. The homemade cardioversion machine provided a sustained electrical current through the heart.

Electrocutions in a residential home are usually accidental and usually occur because of contact with faulty electrical appliances/equipment, faulty electrical wiring, or tampering with electrical outlets. Electrocutions from homemade devices are less common. A past example of accidental electrocutions from a homemade apparatus involved electric worm rod probes; a metal

rod connected to a source of electricity and inserted into the ground to drive worms to the surface. Electrocutation deaths have been reported with homemade electrical contraptions used for autoerotic purposes.^{1,2} Very rarely, homemade devices resembling electric chairs have been used to commit suicide.³ According to this research, this case is the first report of an electrocution death due to a homemade defibrillator.

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Electrocution Deaths, Cardioversion, Atrial Fibrillation

D8 The Internet-Use Characteristics of the Undergraduates in Istanbul and Gaziantep Provinces

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After attending this presentation, attendees will better understand the internet-use habits of undergraduates in Turkey.

This presentation will impact the forensic science community by sharing the interesting and crucial results of the study.

Internet use continues its rapid advance and social penetration, becoming ever more popular worldwide. However, the proliferation rate of internet use in Turkey remains low. In 1998; the number of internet users was approximately 130,000; in 2005, this number has grown to 14 million. Recently, internet use in Turkey has constituted 4.6% of overall European internet use. Internet use in society is complicated, including the determination as to the helpful or negative effects on the public and particularly on teenagers in such a dynamic environment. Recently, the number of undergraduates using the internet has increased sharply. One of the main reasons for this phenomenon is the use of the internet with personal computers, which provides the opportunity to access many Web sites and communicate with a variety of peers. The increase of users and the time spent on-line have revived the question, "Can excessive time on the internet cause negative effects for adolescents?" This research endeavored to assess the prevalence of internet addiction among college students who study on four different universities in Gaziantep and Istanbul.

A total of 1,470 participants were asked to complete the Diagnostic Questionnaire with 30 questions based on an inventory that included socio-demographic factors and questions about their use of the internet and social media. Samples were composed of 55% (804) female and 45% (658) male participants aged 18-45 years old. The sample group was mainly comprised

of 20-year-olds at 24.5% (347) and 21-year-olds at 22.2% (315). The average daily and weekly time on-line was determined as ± 3.3 hours (daily) and ± 20.8 hours (weekly), respectively. Forty-seven percent of participants (897) accessed the internet using their personal computers while 34% (645) accessed the internet with their personal mobile phones. Of the individuals using personal computers, 37.2% (916) of the samples have their own personal computers and, interestingly, all of the participants have accounts on various social networks. According to the types of internet usage distribution, 37.2% (916) of the participants use the internet only for social media tracking. Statistical comparisons of demographic and internet-use characteristics between female and male participants demonstrated male participants are more prone to watch sexually oriented materials, establish new relationships with the opposite sex, and spend more time on-line. Another study about the average weekly internet-use time of 531 students demonstrated an average of 8.1 hours. In this study; the average weekly time on-line was determined as 20.8 hours. Given the results of this study, it appears reasonable to suggest that the high availability of the internet and the increase of time spent on-line may create a risk for possible internet misuse by undergraduates.

Internet Use, Undergraduates, Turkey

D9 Brazilian Serial Killer: A Case Study of the Emasculated Boys

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After attending this presentation, attendees will gain insight on a succession of murderers involving children which occurred in Brazil and will better understand the operation of a police investigation and its impact on the sphere of the criminal justice system. This presentation will also point out the legislative, social, legal, and procedural repercussions resulting from the traditional methodology used, which could have been avoided with the use of an appropriate technique.

This presentation will impact the forensic science community by discussing the value of an interdisciplinary investigation approach and by proposing an alternative way to investigate serial murderers. This presentation discusses two techniques of forensic investigation used to solve an actual case involving serial murderers, "The Case of Emasculated Boys," which occurred in the north region of Brazil between 1989 and 2003. During the course of investigation, it became clear that the results obtained using the traditional methods of homicide investigation were inadequate and aggravated the situation, unlike the results obtained when using the proper techniques for serial crimes.

The traditional way of investigative thinking used by the Brazilian law enforcement is useless in serial crimes, since the criminal motivation, in this case, was not logical, but symbolic and subjective. Through this case study, discussion will involve the major problems occurring during the traditional investigation of serial. This study also intends to clarify the material points for an investigation on serial crimes, such as reliable surveys and interviews. Inconsistent interviews lead to rework during the procedure, which can ultimately be the cause of reasonable doubt, even when substantive evidence is available. Forensic and objective evidence combined with the use of proper scientific tools are necessary to obtain a solid end result at trial.

A research method was conducted through the analysis of cases, employing official law enforcement as well as federal court documentation. A total of 30 cases in Maranhão and 12 in Pará were re-analyzed. Interviews were conducted of personnel involved in all spheres of the cases, as well as analysis of the interrogation and interview with Francisco das Chagas Rodrigues

de Brito, responsible for 42 murders in both states. About 40 hours of interviews were conducted during this research. The benefits of this methodology are speed, low cost, and little need for specific expertise in areas such as psychology, sociology, and criminology. Chagas confessed and has been tried for the murder of 13 boys up to this point, being convicted for a total of 250 year in prison so far.

The work conducted resulted in a significant gain in experience, later applied to several serial murder investigations. In each of them, the sequence of problems was the same: similar crimes which had never been connected as being of the same authorship until there were a significant number of them, not justifying a specific line of investigation. Most of the crimes also involved poor victims or victims belonging to minority groups.

As a result of this research, the best recommendation and motivation for changes in the traditional methodology is the scientific demonstration of the results obtained through the utilization of new tools. It is important to point out that these crimes occurred in poor regions of Brazil, where state-of-the-art forensic tools are not available. It is fundamental to stimulate studies that may increase the understanding of all the problems of high social significance caused by the inadequacy of not using updated investigative methodology.

In the course of this case study, it is proposed that in serial murderer investigations conducted in Brazil, the same sequences of problems are recurrent: inappropriate research methodology; inadequate media coverage and impact; loss of efficiency of criminal justice; and a lack of an interdisciplinary approach resulting in uncoordinated information collection. Therefore, law enforcement and forensic professionals must have access to the data and specific methodology employed on the investigation of all crime scenes that may be connected, increasing the chances of solving such peculiar crimes. Criminal profiling has already been used in other specialized investigations of serial crimes in Brazil, always with positive results.

Brazil, Serial Killer, Homicide Investigation

D10 Volatile Organic Compounds — From Science to Victim Recovery Canine Training: A Method to Aid in Determining the Location of Human Remains

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The goal of this presentation is to demonstrate to the forensic science community the specific Volatile Organic Compounds (VOCs) that elicit an appropriate response from victim recovery canines.

This presentation will impact the forensic science community by illustrating how the identification of these VOCs from blood may be useful to develop specific human aids for the training of cadaver dogs and may eventually allow them to identify human remains.

Because canines rely on olfactory rather than visual cues, a properly trained "cadaver dog" can be effective at locating human remains that may be buried or hidden from view.

From a crime scene perspective, the use of trained canines is also considered "nondestructive." Detection dogs are the most common and widely accepted biological detectors due to their proven relative accuracy, their sensitivity and selectivity, and their ability to be rapidly deployed and cover a large area.

Particular research efforts have focused on establishing the volatile chemical signature of compounds that could indicate the presence of buried human remains where surface clues are lacking.

In fact, human scent is defined as the most abundant VOCs that are identified in the headspace of a collected scent sample; but only a few VOCs, evolved from a biological specimen, can stimulate canine olfactory alerts in every cross-matching condition.

A study was performed to release the VOCs in the human cadaveric blood which stimulate canine olfactory using Gas Chromatography-Mass Spectrometry (GC/MS). The VOCs released into the headspace of four different samples of cadaveric blood (two male and two female subjects, both White and Black) were analyzed.

The inclusion criteria for the selection of the samples, taken from a cadaver, were from cadavers between 20 and 25 years old with multiple causes of death, excluding cases of intoxication.

The cadaveric blood samples were stored at -20°C; but, prior to analysis, the samples were allowed to equilibrate for 21 hours at room temperature (22°C and 45% relative humidity). Then 0.5mL of each single sample were stored in 10mL sample vials and sealed with Teflon®-faced septa caps. Serial injections were carried out at an initial column temperature of 50°C, reducing progressively up to a temperature of 30°C. An injection of methanol was used after each injection of cadaveric blood sample.

Over 100 VOCs have been identified, but some of these are the result of a cross-reaction between the cadaveric blood and environmental contamination. Specific compounds have been identified as key markers of cadaveric samples.

The purpose of this research has been to identify the type and relative concentrations of VOCs present in human cadaveric blood exposed at different temperatures. These results could be included to aid in training canines in order to improve the ability to use olfaction to locate human remains.

Additionally, field trial experiments to determine canine interest in the observed VOCs will be conducted to identify correct canine positive responses (PPV) to these few VOCs, with minimal false positives or false negatives by using the same methodology applied in previous work. There is ongoing university research directed at the development and improvement of victim recovery dogs' performances.

Victim Recovery Canines, Volatile Organic Compounds, GC/MS

D11 A BPA Approach to the Shroud of Turin: A Preliminary Examination of the Left Forearm to Reconstruct the Crucifixion Practice

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After attending this presentation, attendees will understand how a Bloodstain Pattern Analysis (BPA) could be performed on the Turin Shroud to reconstruct the original position

of the man impressed on the linen.

This presentation will impact the forensic science community by demonstrating the potential of the BPA approach on uncommon pieces of evidence, excluding the traditional crucifixion position with arms stretched out on the crossbeam, and suggesting further evaluation of one of the most controversial and valuable Christian relics.

Following what was proposed previously by the American Academy of Forensic Sciences' (AAFS) community, the presumed patterns of blood stains from the crucifixion wounds on the linen of the Shroud were approached from a forensic point of view in order to reconstruct the arms and body position during blood flow.¹ The goal was to better understand how this ancient death penalty practice — of which almost nothing is known — was performed.

Reddish stains are evident in the head area, in the wrist and forearm, on the chest, and on the feet of the image on the Shroud. The present study is focused on the analysis of the left forearm, where all the traces are visible from the wrist to the elbow. The preliminary goal of a more extensive project is to reproduce the most similar stain pattern from a dripping point on the wrist in different arm positions.

A ballistic angle finder was used, measuring the arm-body (or forearm-body) angle from 0° (arm parallel to the ground and perpendicular to the body) to 90° (vertical arm). The end of a transfusion cannula was fixed at Destot's space to simulate the dripping from a puncture-type injury where it is usually believed that the nail for the crucifixion was positioned.

However, since the wrist stain is too large to identify a clear injury spot, two other series of experiments were performed with the aperture of the cannula a little closer to the knuckles or to the wrist.

Whole human blood with anticoagulant was used, the latter having no influence on the flow direction. A transfusion bag was attached to the cannula and placed above the aperture; a rolling clip on the tubing allowed control of the blood flow rate.

All the tests performed clearly demonstrate that the angle between the arm and the body must be greater than 80° in order for the rivulets to flow from the wrist toward the elbow, as it appears on the Shroud. This is more reasonable considering the position of the sentenced person when attached to the cross. Results of this study also preclude the use of any kind of ligature to tie the arm or the forearm horizontally to the crossbeam (*patibulum*) for the "Man of the Shroud." Considering these results, the imprint on the Shroud does not correspond with the traditional artistic image of a crucifix with arms stretched out on the crossbeam; a position with the arms folded backward at the elbow and bent around the crossbeam, as in the painting of Mantegna (*Crocifissione*, 1457-1459) is also not supported by this interpretation. Further analysis will focus on the position of both arms to detect if there is symmetry and to reposition the "Man of the Shroud" on the cross in the most suitable crime-dynamic reconstruction. The final step of this investigation will compare the other reddish stains on the linen (head, chest, and feet) to investigate their correspondence, mutual agreement, and the possible time sequence of their occurrence.

According to the direction of the blood dripping on the left forearm, this study demonstrates that the traditional image of a crucified victim with arms stretched out on the crossbeam perpendicular to the body is not supported for the "Man of the Shroud," who should have been fixed with the arms outstretched upward (arm-body angle greater than 80 degrees).

Reference:

1. Jackson JP et al., The Shroud of Turin as an Object of Forensic Science Investigation. Proceedings of the American Academy of Forensic Sciences; 2010, Seattle, WA.

BPA, Crucifixion, Shroud of Turin

D12 Identification and Management of the West, TX Explosion Victims

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The goals of this presentation are to act as a case study of the West, TX fertilizer plant explosion and to discuss the importance of communication between agencies.

This presentation will impact the forensic science community by giving an example of multiple agencies working together to identify mass disaster victims in a unique death inquest setting. It will allow others to consider possible variations to their own mass disaster plans.

A mass disaster event is always a possibility for any medical examiner's or coroner's office and requires a combined effort by many agencies and individuals. A key factor in any large fatality incident is identifying those among the deceased in order to ascertain if there are any victims still unaccounted. This presentation will describe how the Dallas County Medical Examiner's (DCME) office and McLennan County Justice of the Peace (JP) collaborated to identify the victims of the West Fertilizer Co. ammonium nitrate plant explosion on April 17, 2013.

The West, Texas explosion caused the largest mass fatality of first responders in the country to date, since the fall of the World Trade Center towers in 2001. As a result of the death inquest structure in Texas, the McLennan County JP contracted the DCME office to identify the individuals, perform the autopsies, and other associated services. The situation was unique, for the crime scene was outside of the DCME's jurisdiction and the magnitude of the disaster was unrealized for some time due to the instability of the explosive chemicals, the lack of information about the number of people known in the area, and the number of volunteer emergency responders involved at the time of the incident. A large combined effort from law enforcement agencies, government agencies, and individuals was required to coordinate logistics and relay information.

The victims arrived at the DCME in three different intervals due to access of the blast site and search and recovery efforts. The first two victims were civilians, found in the rubble of an apartment complex near the fertilizer plant. The second wave of victims recovered from the blast site included all 11 first responders and one civilian. Lastly, another civilian from a nearby nursing home was sent two days later, after the JP learned of his injuries sustained during the blast shortly before his death. The first 14 decedents were all unidentified upon arrival to DCME. The original two civilians were noted to have relatively minor external injuries and were identified via fingerprints within four hours of their autopsy. The 12 decedent remains from the immediate blast site were noted to have various degrees of trauma among them, ranging from blunt force injuries to extreme thermal injuries. At the time of the second shipment of victims, a list of names for possible decedents was received with no other information. Within 17 hours of intake, all but two of the victims were able to be identified by fingerprints. Four days later, most of the residents of West, Texas had been located, minus a few who had been known to be in the blast radius. The recovery teams located five more human remains at this time and they were sent to DCME for identification.

Dental records and DNA family reference samples for the final two unaccounted-for first responders were sent five days after the explosion. The remaining two victims and all associated human remains were identified and related within one day of their receipt, via DNA. From the initial arrival time to DCME, all decedents and associated human remains were identified within six days.

This experience demonstrated the tremendous effort and cooperation needed between agencies during a mass disaster. The identification of mass disaster victims is a challenging task in itself. Adding the aspect of relying on another agency to collect family reference data and identification materials, the need for inter-office communication and implementing a thorough but simplistic mass disaster identification system in a medical examiner's office was magnified.

Mass Disaster, Identification, West, TX

D13 Body Rendering by Drug Cartels in San Diego: Case Study of Caustic Soda

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After attending this presentation, attendees will have a better understanding of the effects of caustic soda (sodium hydroxide, lye) on human remains.

This presentation will impact the forensic science community by presenting unusual case circumstances rarely seen in the medical examiner's office.

Ever since Adolph Luetgert tried to render his wife's body in a vat of caustic potash in his Chicago sausage factory (1897), individuals seeking to avoid discovery of a murder or to confound identification have used caustic substances to destroy human flesh and bones. Usually, the substance of choice has been an acid: muriatic acid; sulfuric acid, or hydrochloric acid. Research and case work have shown that these strong acids are very effective at destroying soft tissue and demineralizing bone. Many of these same studies have shown that sodium hydroxide and other alkaline lye substances have minimal effect on tissue integrity. This presentation (as well as Luetgert's experience) demonstrates otherwise.

The case study comes from San Diego County. Los Palillos are a cartel-like gang along the border with Mexico that makes its money by trafficking drugs and kidnapping victims for ransom. They often resort to murder to take out competitors. Between 2004 and 2007, they were responsible for at least nine murders in the San Diego area. Bodies were tossed in streets, left in cars, and, on at least one occasion, dissolved as far as possible, dumped, and buried. A cooperating witness detailed the events for the prosecution.

In May 2007, two men were kidnapped, held for ransom at a residence in Chula Vista, CA, for two weeks, then beaten, strangled, stripped, and dumped headfirst into barrels of water and *sal caustica*, which the cooperating witness had been sent to buy. The perpetrators referred to the substance as "acid" but it clearly was an alkaline lye substance. The barrels were heated for hours, and then the bodies were left to soak for three days. The barrels were transported to a ranch in the Tijuana River Valley, where their contents were dumped into a prepared pit and covered over.

In October 2009, the remains were recovered and brought to the Office of the Medical Examiner for San Diego County for analysis. The remains were received in 55-gallon and 30-gallon drums and two boxes. The boxes contained numerous bone fragments and fourteen teeth. The drums each contained an

odorless semisolid gelatinous mass dotted with bone fragments and flakes of caustic soda. The recipients (a forensic anthropologist and a forensic pathologist) had never seen anything quite like it. Many hours were spent cutting through this greasy mass and cleaning the bone fragments. The bone fragments were basically husks, the protein matrix having been destroyed. This was a very different outcome from what had been seen in previous known cases of acid use, where the bones become quite flexible. A detailed discussion of the nature of the remains will be presented.

The foot bones turned out to be well represented, to the point where three talus and three navicular bones (two lefts and one right) could be identified, indicating two persons. A fragment of ischiopubic bone indicated a male in his late twenties to early thirties, and shoveled incisors suggested possible Southwest Hispanic ancestry. A DNA expert at the crime lab was able to extract usable DNA to identify one of the victims as one of the men who had gone missing two years earlier.

The case went to trial in the spring of 2012. Testimony of the cooperating witness was graphic and compelling. The forensic anthropologist was able to explain to the jury the process of bone sorting and identification. Both defendants were convicted and sentenced to life in prison.

Body Rendering, Lye, Bone Analysis

D14 Hyperspectral Remote Sensing of Individual Grave Sites — Cadaver Decomposition Chemical Effect on Spectral Vegetation and Soil Changes

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After attending this presentation, attendees will gain an understanding of airborne hyperspectral remote sensing as it pertains to locating new and recent clandestine grave sites including known limitations and areas in need of further investigation. The presentation will also cover the effects of cadaver decomposition chemicals on the spectral signatures of soils and vegetation, the changes which occur over time, and how these results can be utilized for detection of clandestine and unknown grave sites.

This presentation will impact the forensic science community by outlining a potential nondestructive tool for the detection of recent individual grave sites using airborne platforms and through relatively rapid assessment to narrow down search areas and prioritization, enabling law enforcement personnel or Search and Rescue to cover a wider area than is possible using ground-based detection methods such as ground-penetrating radar and cadaver dogs.

The use of hyperspectral remote sensing for the detection of clandestine graves is emerging as a potential alternative tool in forensic investigations. Previous studies have demonstrated that it is possible to use hyperspectral remote sensing techniques to detect both mass and single graves. With this multi-year study, the goal is to demonstrate the feasibility of utilizing this same technology for the detection of recent individual burial sites under a wider range of conditions.

Detection of clandestine burials is of interest to police and first responders, with cases arising from victims of crime as well as situations such as missing hunters and hikers where no foul play is suspected. Finding buried bodies can be crucial to solving cases, as well as for providing closure for victims' families. Airborne hyperspectral remote sensing enables coverage of a

wider area than is possible using ground-based detection methods. However, as with all detection technologies, it has its limitations and a fundamental aspect of using this technology for single grave detection is to understand what these limitations are.

Detection is based on the alteration of the environment by the body through decomposition, essentially a form of environmental contamination which can affect both the soil and the vegetation. It is known that a decomposing body alters the surrounding environment and that the changes in the soil matrix can alter plant chemistry. The degree to which this alteration takes place is highly dependent on season, geographical location, vegetation type, and the state of the body when it is buried, as well as characteristics of the body such as the weight. Due to the chemical changes in the soil, plants undergo a stress response, changing the levels of plant pigments. These changes in plant pigments have been shown to be detectable by hyperspectral sensors. The detection of single graves poses a difficult detection problem primarily because the body mass is relatively small and the effects of the decomposition will be limited spatially. Furthermore, several environmental effects may also induce similar spectral responses. However, these responses are generally on a larger spatial scale.

For this study, 20 pig (*Sus scrofa*) carcasses were utilized as proxies for human cadavers. This research examined the effects of three burial scenarios — surface, 30cm, and 90cm soil cover (all with and without the bodies being wrapped in garbage bags) — on the detectability of single bodies (180-200lbs each) from an airborne sensor as well as from laboratory analyses of the spectral signatures of the soil and vegetation. An aircraft equipped with hyperspectral sensors covering the visible-to-shortwave infrared range (450-2500nm) sensors collected imagery as time and weather permitted over the course of three years (2011-2013). In addition, a FieldSpec® 3 Portable Spectroradiometer was used to collect vegetation spectra in the field and soil spectra in the lab.

Soil samples were collected three times monthly during the first year, with two samples in the growing season collected in the second and third years. These underwent aqua regia digestion for element analysis. Chlorophyll and carotenoid values were extracted from the vegetation samples to quantify vegetation pigment differences between background and disturbed soil vegetation.

This study found that burial depth plays a significant role in the detectability of individual bodies based on changes in the soil and vegetation that were especially noticeable shortly after burial. The majority of the spectral changes that were detectable through airborne and handheld sensors in the first year indicated an overall disturbance effect rather than a direct effect on the soil and vegetation from the decomposition chemicals from buried remains. In the second and third years, the effects of the cadaver decomposition chemicals were more apparent in the spectral changes.

This presentation will highlight what this study means for using this technology and will illustrate situations in which these tools are feasible for improving detection rates of clandestine graves as well as describe the detailed calibration procedures that are required in order to adequately use such data to locate the graves.

Hyperspectral Remote Sensing, Cadaver Detection, Clandestine Graves

D15 Comparison of Extraction Methods: Extracting Explosives From Soil Matrices

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After attending this presentation, attendees will have a basic understanding of effective ways to extract explosives from soil matrices. More specifically, attendees will become familiar with the differences between using ionic liquids, common solvents, and the Environmental Protection Agency (EPA) method 3535A to extract explosives from soil.

This presentation will impact the forensic science community by providing a comparison of extraction methods for explosives in soil matrices. To date, there is not one single commonly used method to extract explosives from soil. Most of the current extraction processes take ample amounts of time to complete, and/or can yield inconclusive results. This study will evaluate and determine the most efficient extraction method presented in comparison to currently used methods.

Terrorism is a common threat throughout the world. Many terrorist attacks involve the use of secondary explosives and, when detonated, can leave trace amounts of explosives in surrounding soil matrices. Currently, there are many different methods that forensic scientists utilize to extract explosives from these soil matrices, most of which are time-consuming and dependent on the analytical technique being used.

While many studies have explored the use of one specific extraction method with one analytical technique, this study explores three separate methods to extract explosives from soil matrices in order to see which method is more sensitive, effective, and cost beneficial. In addition to using three different methods of extraction, three separate analytical techniques were utilized for each extraction method.

For this study, RDX and TNT, two commonly found secondary explosives, were added to two different types of soil matrices. Each soil sample was then run through each of the three following extraction methods: use of ionic liquids; saturation in common solvents; and EPA method 3535A. When using the ionic liquid method, the explosive containing soils were completely saturated in ionic liquids or the ionic liquids were placed on a swab that was run across the top of the soil. Ionic liquid samples were then run through Solid Phase Extraction (SPE) before being analyzed. For the common solvents method, soil samples were fully saturated in a solvent, and without going through any further processes, were analyzed. The EPA 3535A method saturated the soil samples in water, then the soil samples were run through two different SPE methods prior to analysis.

Once each extraction method was complete, the samples were analyzed for the presence of RDX and TNT with the following three analytical techniques: Direct Analysis in Real Time (DART®); Gas Chromatography-Mass Spectrometry (GC/MS); and Liquid Chromatography-Mass Spectrometry (LC/MS). DART® was utilized because of its fast turn-around time; however, because it is only a screening method, both GC/MS and LC/MS were used as confirmatory tests. At the moment, GC/MS is the most common analytical technique used for explosives in a forensic laboratory. However, LC/MS is becoming more prevalent in labs and has demonstrated greater sensitivity when analyzing chromatographic data.

After all three extraction methods were utilized and samples were run on the DART®, GC/MS, and LC/MS, the

chromatographs were analyzed. A comparison was carried out to determine which extraction method was most beneficial based on the chromatographic results, extraction preparation time, and analysis time, combined with the overall cost of each method.

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Extracting Explosives, Extraction Method, Explosive Analysis

D16 The National Ballistic Imaging Comparison Projects

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After attending this presentation, attendees will learn: (1) how the use of reference materials; (2) control charts and limits; and, (3) attendant statistical analyses that characterize the variability of measurement processes involved in image acquisition for some ballistic identification systems.

This presentation will impact the forensic science community by presenting case studies of how measurement quality studies using reference materials helped to maintain and improve the measurement process in ballistic laboratories.

When evidence bullets or casings are found at a crime scene in the United States, the National Integrated Ballistics Information Network (NIBIN) can be searched to find the bullets or casings most similar to the evidence bullet or casing that has been previously entered into the database. Spent casings usually have three areas that contain tool mark signatures left by the following parts of a firearm: the firing pin, breech face, and ejector. The surface of a fired bullet often contains Land Engraved Areas (LEAs) that are tool marks engraved (scraped) on the bullet by the rifling inside a gun barrel. Images of these important areas are acquired and entered into the NIBIN system. Comparisons with ballistic impressions from known firearms and evidence already in the database are carried out utilizing a proprietary similarity or "correlation" score between the relevant images.

The value of such ballistic database searches is dependent on the quality of the measurement and imaging process. The National Institute of Standards and Technology (NIST) has developed a standard bullet (NIST SRM 2460) and a standard casing (NIST SRM 2461) to help forensic laboratories maintain and improve their measurement procedures. The standard bullets and casings are manufactured to resemble tool marks found on actual bullets and casings and have been certified to be almost identical to a reference standard bullet and casing, respectively. The surface of the standard casing contains electroformed marks that replicate signatures impressed by a firing pin, breech face, and ejector. The surface standard bullet contains marks that are diamond-turned

replicates of six different LEAs.

The first National Ballistics Imaging Comparisons (NBIC-1) project involved a select group of volunteer operators of NIBIN instruments from across the country. Each was given access to a standard bullet and a standard casing and instructed to perform a series of periodic image acquisitions of their bullet and casing. The goal was to characterize what would be the typical values and variation of the correlation scores of their acquired images as compared to "Golden Images" of the same areas of the standard bullet and the standard casing that had been previously entered into the NIBIN system. The NBIC effort was not meant to be a survey of then-current operator practice. Rather, it was intended to be both diagnostic and exploratory, with the ultimate goal being a baseline of best practices, formalized by control limits, with control charts for monitoring individual performance. Thus, the fruits of NBIC-1 included not only control limits for each ballistic area measured (based on data from the operators), but also some improvements in the performances of operators, the system, and the standard casings.

In the last few years, the image-acquisition instruments for casings used in NIBIN are being replaced by a newer version of the instrument that, among other things, automates part of the image-acquisition process. The second National Ballistics Imaging Comparisons project, NBIC-2, involved another group of NIBIN operators performing image acquisition of the standard casing on the newer instruments. The goal is to characterize and compare the ensuing typical values and variation of correlation scores, including revised control limit values. In addition, some insight into differences between the two systems and their respective "Golden Images" is obtained.

NIBIN, Firearm, Ballistic Identification

D17 The Phenomenon of the Urban Mummy

Barbara C. Wolf, MD, District 5 MEO, 809 Pine Street, Leesburg, FL 34748; and Brett E. Harding, MBA, District 5 MEO, 809 Pine Street, Leesburg, FL 34748*

After attending this presentation, attendees will have an understanding of the constellation of environmental factors and events leading to the mummification in an urban setting and the importance of the multidisciplinary approach to the medicolegal investigation of such deaths.

This presentation will impact the forensic science community by contributing to the increased awareness of the study of these unusual cases and the unique aspects of the medicolegal investigation of these deaths.

The environmental mummification of a body in an urban setting is a fairly unusual event. Florida is known for its high temperatures and humidity, which typically contribute to the rapid decomposition and putrefaction of decedents' bodies. The resulting foul order is frequently the signal leading to the discovery of the bodies of individuals whose deaths occurred in solitary locations. This, coupled with the prevalence of insect activity, usually precludes the mummification process. However, under certain circumstances, Florida's unique climate provides an environment suitable for mummification. A particular constellation of human behaviors and scenarios must coincide for this phenomenon to occur in an urban setting.

The first requirement is that the decedents must have lived in socially isolated circumstances. Such isolation may result from multiple etiologies, such as illness, substance abuse, criminal activity, language barriers, and/or advanced age. Florida is home to a large number of individuals who lead an isolated existence. The homeless, who may purposely isolate themselves as a safety

measure to protect themselves and their property, the mentally or physically ill, who are isolated as a result of their impairments, and the elderly, whose longevity may result in their isolation after the deaths of friends and family, are all at risk. Reclusive behavior is almost essential for a death to go unnoticed and to meet the time requirements for the decompositional mummification process to proceed.

The apathy of nearby individuals to the welfare of the decedent or the lack of knowledge of the decedent's presence in the community is another factor. If neighbors call attention to the absence of the decedent, a well-being check will usually interrupt this process. The failure of public officials to recognize the signs of a potentially undiscovered death is also a necessary condition. These may include law enforcement, mail carriers, and/or public safety officials. Even with isolationist behavior, violations of urban laws or ordinances may result in the inspection of a home by public officials. These public service representatives must be apathetic to the absence of the decedent or equate his or her absence with the abandonment of the dwelling. Finally, the body must be in a hot, dry, closed environment where it is protected from insect and animal predation. The death of an individual in an exposed location is often appreciated by the local fauna as a ready food source long before humans become aware of the death. This predation results in the consumption and scattering of the remains rather than their mummification.

This research recently investigated several cases in which mummified bodies were discovered in urban locations in southwest Florida. The literature pertaining to such unusual cases is sparse. These cases highlight the importance of the multidisciplinary approach to the medicolegal investigation of such deaths and the determination of the cause and manner of death.

Mummification, Urban Setting, Multidiscipline Investigation

D18 Determination of Autoerotic Asphyxial Deaths

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After attending this presentation, attendees will gain an appreciation of common misconceptions surrounding autoerotic asphyxial death scenes. Identified will be ten criteria and two exclusions that death scene investigators can utilize to promote increased proficiency in investigative actions in potential Autoerotic Asphyxiation (AeA) scenes. Five cases of this secretive and potentially deadly sexual practice in the Central Florida region are retrospectively evaluated from the records of a county medical examiner's office using the principles prescribed.

This presentation will impact the forensic science community by emphasizing that the manner of death classification is important because deaths resulting in the practice of AeA are accidents as opposed to asphyxial deaths without AeA components, which are classified as suicides. Additional considerations inherent in AeA scenes can involve the disbursement of insurance claims and psychological issues of the autoerotic practitioner, making this a crucial forensic science matter. The significance in identifying these objectives will not only provide recognition of AeA, but will also assist in manner-of-death determination by assisting law enforcement and medicolegal experts in evaluating AeA cases. Additionally, it will provide insight into the psychological aspects of practicing individuals. Furthermore, the information will assist law enforcement and medicolegal experts in evaluating AeA cases.

The activity of AeA is often revealed at the medical examiner's office or the coroner's office due to a faulty safety mechanism in the asphyxial device leading to the practitioner's demise. AeA shares common elements with asphyxial suicides and is often interpreted as such. The intention of the participant in this act is not for a fatal outcome; on the contrary, it is to achieve heightened sexual gratification.

Unfortunately, there are unique circumstances in AeA death scenes that may mislead the investigator. These include loved ones tampering with the death scene for the purposes of minimizing perceptions of social disgrace and the non-recognition of assorted paraphilias associated with AeA that rule out suicide. It is important for the investigator to recognize an altered death scene.

This study elucidates ten novel characteristics involved in AeA that will differentiate it from an asphyxial suicide scene. It will also assist in manner-of-death determination, identify paraphilias associated with AeA at death scenes, and demonstrate that the act is often repetitive with an increasing level of complexity as the practitioner matures. It is recommended that medicolegal death investigators become familiar with these AeA criteria.

Autoerotic, Asphyxiation, Hypoxyphilia

D19 Toward a Better Understanding of Sexual Violence Among Female Youth Gang Members

Cliff Akiyama, MPH, MA, PCOM, Dept of Forensic Medicine, 4170 City Avenue, Philadelphia, PA 19131-1694*

After attending this presentation, attendees will be able to identify the signs and symptoms of intimate partner violence in the female youth gang population. They should also be able to recognize some of the barriers to getting help and, most importantly, know where to turn for assistance if they suspect intimate partner violence in the female youth gang population. Moreover, the attendees will be able to analyze some novel prevention/intervention programs that have worked in the community to help stop this problem before it's too late.

This presentation will impact the forensic science community by informing them of an extremely hidden topic of sexual violence in the female youth gang population.

Youth gangs throughout the United States continue to rise as well as terrorize the neighborhoods that they claim as their own, causing the citizens in these gang-infested neighborhoods to live in constant fear of their lives every single day. According to the Office of Juvenile Justice and Delinquency Prevention of the United States Department of Justice, there are over 24,500 gangs in the United States with a total gang membership of 750,000. Often overlooked in gangs is the pervasive bondage of sexual violence among female youth gang members. In Los Angeles County, California, alone, there are currently 9,000 female gang members, while in Philadelphia County, Pennsylvania, there are 430 female gang members. Demographics show across both counties a female gang member average age of 15 years with a range of 8-22 years. Over 400 female gang members were interviewed in the streets, jails, and juvenile halls, using a target questionnaire in Los Angeles and Philadelphia. This study found that over 80% of all female youth gang members are "sexed in," as an initiation tactic of getting into a youth gang. Getting "sexed in" the gang means that the prospective female gang member has to have sexual intercourse with all of the male gang members in that particular gang set or clique. Moreover, the study found that 25% of the female gang members interviewed have a history of child sexual abuse. Unfortunately, female gang members are

least likely to report rape victimization due to fear of death by the male gang members. The study also found that of the female gang members that are physically and/or sexually abused by the male gang members within the gang, 97% have never utilized any rape or physical abuse services such as a rape crisis center or shelter within their communities, while 60% of the female gang members do not even know about the services available to them. Furthermore, with the technical assistance of Women Organized Against Rape (WOAR), a Philadelphia, Pennsylvania, rape crisis center, this study identified 12 distinct barriers to disclosure and service utilization of female youth gang members who experience sexual and physical violence across their lifespan (i.e., immigration issues, racism, depression, anxiety, fear of death, shame, isolation, age, lack of health insurance, lack of child care, lack of affordable housing, lack of independent income, lack of support from the family/community, cultural fluency, and just not knowing the available resources surrounding intimate partner violence).

Youth Gangs, Sexual Violence, Youth Violence

D20 “Lover Come Back:” Rituals, Resurrection, and Cultural Beliefs and Their Impact on Death Investigation and Grieving

Rae H. Wooten, BSN, RN, 4050 Bridge View Drive, #500, North Charleston, SC 29405*

After attending this presentation, attendees will be able to list at least three areas of concern associated with deaths involving those with strong religious and/or cultural beliefs and discuss some potential ways of addressing them.

This presentation will impact the forensic science community by identifying potential challenges facing death investigators when the death involves those with strong religious and/or cultural beliefs, thereby allowing for anticipation, planning, and management should difficulties occur. It is important this be done in a way in which the investigation is not compromised and those grieving are not subjected to more pain and suffering.

We live in a world where modern travel has facilitated the comingling of endless numbers of ethnic groups with various cultural and religious beliefs. These very important aspects of our lives also transcend death, impacting survivors and their grieving, as well as potentially impacting the investigation of the death.

This study presents a case in which a 34-year-old male suffered a witnessed collapse while playing basketball and was subsequently found to be pulseless and apneic. Cardiopulmonary Resuscitation (CPR) was initiated almost immediately by an off-duty paramedic who happened to be on scene. Attempts to resuscitate the decedent failed and he was pronounced dead approximately 35 minutes later in the emergency room. After talking with the decedent's mother, who is an RN, his wife, and various friends, no explanation for his sudden death was determined. It was also learned that the decedent had traveled to underdeveloped countries in recent months and had possibly been exposed to various diseases. It was determined that a full forensic autopsy, to include toxicology, would be necessary in order to determine the cause and manner of death.

Very shortly after the death occurred, the decedent's wife communicated that she wanted the body to be transported to a private residence in the area. She forbade an autopsy in the strongest terms possible, threatening court action and litigation. When she, and a number of individuals who accompanied her, arrived at the hospital, they demanded immediate release and access to the body in order that he might be resurrected. All efforts to explain the need for an autopsy were met with increasingly angry demands. It became apparent during many discussions that their

belief that the decedent could be resurrected was based on firmly held religious beliefs.

Death investigation should take into consideration the cultural and religious beliefs of each decedent and their families, and typically neither those beliefs nor the investigation need to be compromised. In this case, no agreement could be reached and having a responsibility to the decedent, the family, and the community, an autopsy was performed over the wife's objections. It revealed a coronary artery thrombosis and this was discussed with the wife. She continued to demand access to the body and after much thought and planning, the body was moved from the morgue to a secure room where the wife and others engaged in ritualistic activity and behaviors for several hours. Eventually, the body was forcibly returned to the morgue in spite of their beliefs that there were signs of life; in their view, the decedent died as result of these actions.

The decedent's parents, siblings, etc., who did not share these beliefs, were very distraught which resulted in a "Temporary Restraining Order" which forbade the release of the body until some agreement could be reached. Funeral services were delayed, resulting in additional pain and suffering for many, legal fees were accrued, and a tremendous amount of time and energy was consumed.

This case points out that in some instances, no matter what steps are taken to be sensitive to and supportive of cultural and religious beliefs, it will be perceived otherwise. It is therefore important to consider ways in which situations such as this might be handled so that beliefs are respected and the investigation is not compromised.

Cultural Beliefs, Resurrection Rituals, Possession of Body

D21 The Cops Collected the Bugs in This Abuse Case — Hooray!

Neal H. Haskell, PhD, 425 Kannal Avenue, Rensselaer, IN 47978*

After attending this presentation, attendees will learn the importance of insect collection, what additional information can be concluded from those insect identifications during a child abuse investigation, and how, from these facts, a jury was able to view the conclusive evidence and render a guilty verdict against the child abuser.

This presentation will impact the forensic science community by increasing understanding of why and how to collect entomological evidence, and the importance of collecting and presenting that evidence in court, which will lead to increased convictions in wrongful abuse cases.

The use of insect evidence to determine the Postmortem Interval (PMI) and answer other questions surrounding a death scene has been in common use in hundreds of case investigations for over two decades around the globe. With this increased use, court appearances by forensic entomologists have been increasingly beneficial to the courts. However, even with this greater recognition of its use, in many jurisdictions across the country there is still an unwillingness to recognize the importance of the entomological evidence, resulting in failure to collect forensically important specimens. The use of entomological evidence may even be declining in these death scene investigations. Whether this is due to the economic environment at this particular time because of lack of funding for training or the perception that expert analysis is cost prohibitive, the truth may lie in this philosophy being "penny wise and dollar foolish." If only the investigators would have collected the insect evidence, one could have had the means to answer some of these most important questions.

In a case from Marion, Indiana, officers were dispatched

to a dwelling where filthy living conditions were reported to meet with Child Protective Services (CPS) personnel. Even before entry into the house, many cockroaches were observed crawling on the outside of the front door as the investigative team entered. Once inside, cockroaches were observed everywhere including on the carpet, clothes, trash bags, freezer, refrigerator, stove, kitchen sink, panty shelves, walls, beds, and even the ceiling. Two baby beds had cockroaches crawling on them in addition to numerous house flies. There were five small children living in this "home," including: a six-year-old boy, a four-year-old boy, a three-year-old girl, a two-year-old boy with special needs, and a one-year-old girl. On initial contact with the children, the 2-year-old was unresponsive and thought to be dead. The children were taken to the local hospital for an examination of their injuries. The three younger children had lesions consistent with cockroach feeding wounds, which were photographed. The two older boys had scars which could have originated from this type of injury. Photographs of the 2-year-old boy were taken to show adult house flies feeding on seeping head wounds, likely from the cockroach feeding. Upon further examination once the diapers were removed, insect fly larvae were discovered in and around the anus. The police officers recognized the importance of these specimens and collected them for further examination and identification. The mother, who was pregnant with her sixth child, and the live-in boyfriend were transported to jail for questioning. There were excellent photographs of the adult flies and cockroaches as well as actual specimens of the fly larvae which were identified as the house fly (*Musca domestica*). Two days before Thanksgiving, the boyfriend was tried first and compelling evidence and testimony was provided by the CPS witnesses, the police investigators, the forensic entomologist, and even the 6-year-old boy. The mother's attorney was present for this trial and, after seeing the testimony against his client, realized there was no way he was going to trial and a plea agreement was reached. Within a week of the children being removed from these terrible conditions, every one of them began to thrive, gaining weight, and showing major improvements to their health. The police and CPS investigators provided the means for additional testimony by taking detailed photos and collecting the larvae so the specific species identification could be accomplished. A conviction was reached for the boyfriend and, since it was his third felony, a 25-year sentence was imposed. The defense attorney said, "We were holding our own until the kid got on the stand, but that killed us." Also, the jury was given a lesson on house fly biology and reminded that, "when they were at the picnic at the park, that little house fly crawling across their potato salad may have just come from a dog pile a few feet away."

Forensic Entomology, Cockroaches, House Fly

D22 Future Forensic Education — An Integrated and Ongoing Approach

Stewart Walker, PhD, Flinders University, Phys Sci, For & Analytic Chem, GPO Box 2100, Rm 304, Adelaide, SA 5001, AUSTRALIA*

After attending this presentation, attendees will be aware of the potential for integrating educational institutes with forensic providers, companies, and government agencies for ongoing forensic education.

This presentation will impact the forensic science community by helping foster a more open attitude toward developing collaborative educational resources.

Like all other areas of education, forensic education is undergoing a dramatic change in the way materials are sourced and presented. Using examples, this presentation will look at what is required for delivering realistic forensic education by

integrating material from educational institutes and from forensic practitioners. Examples will come from a Forensic and Analytical Chemistry degree run at Flinders University, Adelaide, Australia, which combines input from academics, forensic providers, and a range of government and industrial presenters and also from the experience of a Director of the Center of Expertise in Energetic Material, a collaboration between Flinders University and Defense Science and Technology Organization, Australia, and national and international partners.

Advantages of integrating educational material from a range of sources will be discussed along with some of the problems encountered. Overall, the advantages outweigh the disadvantages more realistic material is provided, producing graduates who are better prepared for the workplace

Ideally, students and academics would be placed in working laboratories and the university would host practitioners to ensure a cross-pollination of ideas and the development of procedures; however, limitations of numbers and health and safety regulations may restrict these opportunities.

Examples of successful collaboration will be provided, some of which have led to awards from the Australian National Institute for Forensic Science and the Australian and New Zealand Forensic Science Society.

In all examples to be discussed, the desired outcome is to have education flowing in both directions where the practitioners, supplying their expertise, also pick up new techniques and refresh their knowledge base. In this way, practitioners can refresh their basic skills, upgrade their qualifications, and be up-to-date on the most modern technologies and students can be provided with realistic material and be well prepared to enter the workforce.

Education, Future, Collaboration

D23 Quantifiable Examination of Partial Fingerprints at Crime Scenes: A Digital Technique for Teaching and Field Examination

John Z. Wang, PhD, 18737 W Place, Artesia, CA 90701*

After attending this presentation, attendees will learn the following information: (1) a new digital technique that can perform quantifiable measurements of partial fingerprints; (2) the scientific principles and methodology that support the examination; and, (3) an evaluation of the reliability and validity of the methodology of partial fingerprints for a potential field use.

This presentation will impact the forensic science community by introducing a new digital technique that can examine partial fingerprints with quantifiable measurements in an educational setting and a potential field examination at crime scenes.

Fingerprint examination has been a vital method for inclusion and exclusion of suspects for over a hundred years. However, the field is being challenged in recent years on three major fronts. First, the 2009 National Academy of Sciences (NAS) Report, *Strengthening Forensic Science in the United States: A Path Forward*, challenges the current fingerprint examination to be non-scientific due to its nature of being a pattern- and minutia-based comparison, lacking a quantifiable measurement. Second, the Automated Fingerprint Identification System (AFIS) or the Integrated Automated Fingerprint Identification System (IAFIS), the software for law enforcement agencies in the U.S., is a station-based technology and can provide little support for a preliminary examination at crime scenes. Finally, AFIS or IAFIS requires a certain level of fingerprint quality to be developed at crime scenes. A partial fingermark is not eligible for an examination and comparison

at all. In reality, a field technique for a preliminary examination or at least for a quick exclusion is a timely requirement for crime scene technicians and investigators. This presentation introduces a new digital technique that examines partial fingerprints for an educational setting and possibly for a preliminary examination at crime scenes.

It is argued that to echo the above-mentioned challenges, the 21st-century technology for fingerprint examination should be optical, digital, and portable. The optical technology can provide a non-destructive method and can be used either before or after the lifting of a fingerprint for a safer development. The digital technology tends to be more reliable and valid, making possible a qualitative evaluation and quantitative measurement of full or partial fingerprints. Finally, a new application of digital technology can perform a rapid field examination, providing much-needed support for crime scene technicians and investigators.

This presentation is a preliminary report of a pilot study (N=60 in 30 pairs). The methodology is based on a target sampling approach due to being a pilot experiment. A partial fingerprint can be in the form of a plastic, patent, or latent fingerprint. This study focuses on the latent fingerprint that is made visible after a magnetic powder processing and lifted onto hinged fingerprint cards. The criteria for a partial fingerprint selection are threefold: (1) the fingerprint contains about 25% of the total area; (2) the partial fingerprint is visible by naked eye for an initial visual screening and is readable by a magnifier; and, (3) the partial fingerprint contains at least three minutiae or partial core structure for a triangulation analysis.

For the sake of simple selection of the partial fingerprints, two classifications can be observed and compared: a partial fingerprint with at least three minutiae and a partial with less than three minutiae. The comparison of the 30 pairs is divided into three situations: (1) a full fingerprint is compared with another full fingerprint (F with F); (2) a partial fingerprint with a full fingerprint (P with F); and, (3) a partial with another partial (P with P). The quantifiable examination consists of four types of measurement depending on the location and the type of minutia: linear; area; angle; and ridge-counting comparisons.

The preliminary results of 30 pairs indicate that the success rate is reachable at 95% for the F with F group, 70% for the P with F group, and 50% for the P with P group. It is concluded that the new digital technique is very promising in an educational setting and has a greater potential for partial fingerprint examination for exclusion at crime scenes. It is recommended that the comparison module and interactive graphic interface gives more accuracy, precision, and specificity on fingerprint examination. In order to compare partial fingerprints at crime scenes, a better dactyloscopy with the new digital technique can provide an interactive comparison module with various processing functionalities. In sum, this presentation should be considered as one big step forward to address the challenges from the NAS Report, *Strengthening Forensic Science in the United States: A Path Forward*, which was issued five year ago.

Partial Fingerprints, Crime Scenes, Digital Technique

D24 Medicolegal Deaths Requiring On-Scene Investigation

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After attending this presentation, attendees will understand how the death investigator will be able to identify which death scenes (by suspected Manner of Death/Cause of Death (MOD/COD) type) require on-scene investigation.

This presentation will impact the forensic science community by increasing awareness of the inconsistency across the United States as to which death scenes get on-site investigation by the medicolegal agency (medical examiner/coroner) in charge of the case.

Statement of Problem: Although each state has statutes denoting the types of cases that need to be reported to, and "investigated" by, the Medical Examiner's or Coroner's (ME/C) office, there is variability across medicolegal jurisdictions as to the definition of "investigated." This variability may range from an investigative "review" of basic case information conducted via telephone to an "on-scene" investigation conducted by a medicolegal death investigator for the same type of death. This investigative inconsistency results in medicolegal investigations of inconsistent quality, which can impact the quality of forensic autopsies and death certification. To determine the type of death scene investigation recommended for different (death) scenarios, this study conducted a "case-based" interactive web-based survey of medicolegal death investigators.

Methods: The survey population included active members of the International Association of Coroners & Medical Examiners (IAC&ME), the National Association of Medical Examiners (NAME), the Society of Medicolegal Death Investigators (SOMDI), and those certified by the American Board of Medicolegal Death Investigators (ABMDI) (n=2,392). To identify which case "types" would most likely require "on-scene" investigation, this study developed an automated case "scenario generator" that created unique scenarios based on combinations of six randomly selected categories of case variables (cause and manner of death, witnessed event, age, death location, and body) for evaluation by survey participants. Each category included a set of choices that typically apply to the category (e.g., witnessed: yes/no). After removing combinations that would produce "unlikely" scenarios (e.g., manner=suicide, age=infant, etc.), the six categories and their associated choices (n=45) yielded a total of 9,987 unique case scenarios. Case scenarios were compiled and presented at random to survey participants in systematic groupings of ten cases by age group, each group of ten containing a fixed number of cases by age (one infant, one child, one teenager, four adults, and three elderly).

Results: A total of 468 individual respondents (20% of potential respondents) evaluated a total of 24,352 case scenarios. Preliminary analysis identified six MOD/COD combinations, one body location, and three age categories (e.g., infant to 12 years) that, regardless of all other variables involved in a case, played a significant role in the survey participants' decisions to conduct an on-scene investigation.

Conclusions: Using consensus-seeking methods, standard criteria can be used to determine if on-scene investigation is advised.

Death, Scene, Investigation

D25 Elder Abuse: Following a Felony Case Through the Judicial System

Amy Y. Carney, PhD, 210 Ivory Gull Way, San Marcos, CA 92078*

After attending this presentation, attendees will be aware of the dynamics in investigation and prosecution in felony elder abuse.

This presentation will impact the forensic science community by raising awareness of the process of investigation and successful conviction in felony elder abuse and by focusing attention on the need for an emphasis on personal safety for elders in their homes.

Elder abuse of all types is a growing problem across the United States. As the population continues to age, more cases of felony abuse are being exposed and prosecuted successfully. This case study follows the successful arrest, investigation, and prosecution of two perpetrators in the kidnapping and torture of 76-year-old Natalie "Sandy" Herbst-Vingie.

In 2008, Natalie "Sandy" Herbst-Vingie was a 76-year-old widow, living alone in the Mt. Helix area of La Mesa in San Diego County, California. She was the president of the San Diego Swing Dance Club and was described as a gregarious hostess known for her upbeat attitude. She first met Jeffrey Edward Nelson, age 19, when he came to her door on October 23, 2008, to sell her a vacuum cleaner, which she ultimately purchased. She saw Nelson again on the morning of December 8, when he dropped by to thank her for helping him win a trip to Hawaii in a sales contest. During that visit, a friend of Mrs. Herbst-Vingie came by for coffee and Nelson left. Later that evening, he returned around 10:00 p.m., telling her that he had broken up with his girlfriend and asked to use the phone to call his mother. She let him in, pointed to the phone, and turned away. Nelson attacked Mrs. Herbst-Vingie from behind, choking her. She passed out and when she woke up, she was on her kitchen floor with her knees, ankles, and wrists bound with duct tape. Nelson dragged her to the garage and put her in the trunk area of her car, a 2006 Dodge® Magnum®.

Over the next 26 hours, Mrs. Herbst-Vingie was punched in the face and denied water and bathroom breaks; she was thrown around the trunk of the car as Nelson and an accomplice, Luis Osborne, and a friend of his, Antoinette Baker, drove erratically through the county. She was found and freed when the car was stopped for speeding and running a red light. A jury found Nelson guilty of attempted murder and torture, and he was sentenced to two consecutive life terms in prison plus nine years. He also pleaded guilty to kidnapping and residential burglary. Osborne pleaded guilty to physical abuse of an elderly victim, and second-degree robbery; he was sentenced to ten years and four months in prison.

This case focuses attention on the need for the elderly to be aware of safety in their own homes, as well as the diligent prosecution of felony crimes against older persons.

Elder Abuse, Felony, Conviction

D26 Sexual Dimorphism of Palmprint Ridge Density in a North Indian Population

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After attending this presentation, attendees will understand the usefulness of an important part of palmprints, i.e., sexual dimorphism in palmprint ridge density, and its usefulness in distinguishing gender in the palmprints recovered at the crime scene. The attendees will also learn the specific methodology used in calculating the ridge density in determination of gender from palmprints.

This presentation will impact the forensic science community by presenting the usefulness of palmprints recovered at the crime scene, especially in distinguishing gender using ridge density and may motivate crime scene investigators to endorse this topic for research and practice.

Fingerprints and palmprints are unique to an individual, and forensic scientists have long been using these in the identification of criminals. In the recent past, ridge density (ridge count in a defined area) in fingerprints and palmprints has been investigated especially for its applicability in determination of sex from the fingerprints and palmprints recovered at the crime scene. The present study is conducted to investigate the sex differences in the palmprint ridge density in a North Indian population and explore its applicability in the determination of sex. The study sample consists of 157 healthy young adults (110 females and 47 males) from Shimla city in North India. Bilateral palmprints were taken from all the participants following a standard methodology. The palmprints were manually analyzed in four defined areas of the palmprint. These include central prominent part of the thenar eminence (P1), area proximal to the axial triradius of the hypothenar region (P2), area proximal to the triradius of the second digit in the medial mount (P3), and area proximal to the triradius of the fifth digit in the lateral mount (P4). The ridge density was calculated diagonally on a square measuring 5mm × 5mm according to the method described in literature. The sex differences in palmprint ridge density were statistically analyzed for each of the designated areas using the non-parametric Mann-Whitney test. Differences between right and left sides were analyzed using the Wilcoxon test for paired data. Karl Pearson's correlation was utilized to study the correlation of palmprint ridge density between the four areas on a palmprint. Ridge densities analyzed on different areas of palmprints were compared with each other using the Friedman test for related samples. Total palmprint ridge density was calculated as the sum of the ridge density in all four areas (P1 + P2 + P3 + P4) and analyzed for sex differences. Receiver Operating Characteristic (ROC) curve analyses were done to test the overall ability of the palmprint ridge densities obtained from each area in determination of sex.

The mean palmprint ridge density was found to be significantly higher in females than males in all four defined areas of the palmprint. Ridge densities in P3 and P4 areas of the palmprint show statistically significant bilateral differences (right-left) in both males and females. The study observes variations in the ridge density between the four designated areas of the palmprint. Statistically significant correlation is evident in the ridge density between a few areas among males and in most of the areas among females. Statistically significant differences in the palmprint ridge density were evident between the four areas analysed on the palmprints on the right and left sides among both males and females. Based on the Area Under the Receiver Operating Characteristic (ROC) Curve (AUC), maximum sexing potential for the palmprint ridge density was observed in the P4 area, followed by P3 on both right and left sides. ROC analyses of the total palmprint ridge density indicate that the sexing potential from the right and left palmprint was 70.2% and 72.8%, respectively. The study concludes that the palmprint ridge density can be utilized in the determination of sex as a supportive parameter.

Personal Identification, Palmprint Ridge Density, Sex Determination

D27 Study on Phloxine B Doped Nanocomposite for Fingerprint Detection

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The goal of this presentation is to present a new method to develop latent fingerprints.

This presentation will impact the forensic science community by extending the knowledge of the detection of fingerprints.

Silicon dioxide-based nanocomposites offer large loading capacity for various doping chemicals or molecular complexes, high surface-to-volume ratio, and customizable surface chemistry for the creation and development of novel sensors and devices. When compared with other sol-gel materials, xerogels represent a class of nanocomposites that are relatively easy to fabricate but with unique thermal, acoustic, optical, and mechanical properties for rapid sensor or device prototyping development. When impregnated with fluorescent compounds in their nanosize cavities, the doped xerogels exhibit strong and stable fluorescence properties that are useful for the developing of ion-exchange sensors and optical devices. However, the use of these fluorescently doped xerogels in forensic applications was still largely unexplored.

Phloxine B is a popular fluorescent compound and has shown great application in dyestuff industry as a fluorescent dye to enhance the color of different substrates. But the applications of Phloxine B doped nanocomposite in forensic science, latent fingerprint detection in particular, were still unclear. The use of Phloxine B doped nanocomposite to form a stable fluorescent material for the purpose of latent fingerprint detection was the main goal of this study.

The Phloxine B doped in silicon dioxide-based nanotemplate has been prepared successfully by the sol-gel method. The Phloxine-doped, silicone dioxide-based xerogels were ground into a fine powder using a mortar. Iron powder was added to the xerogel powder with a proper mixing ratio. Labeling powders for latent fingerprints detection do not have to be magnetic. However, the use of magnetic powders with a magnetic applicator offers the unique advantage of avoiding smudging and destruction of fingerprints involving non-magnetic powders and better results on porous surfaces especially. The detection for latent fingerprints formed under different conditions has been studied. The results show that Phloxine B in a Tetraethyl Orthosilicate (TEOS) template exhibits high fluorescence intensity and labeling efficiency. The fluorescence properties of the doped gels in storage or in labeled fingerprints are stable for months in the research laboratory. The study indicates that the Phloxine B-doped xerogel is useful as fingerprint labeling material due to its strong fluorescence properties and photo-stable properties. The Phloxine B-doped xerogel is able to label all forensic-relevant substrates, including aluminum foil, glass, paper, and soft and hard plastic surfaces. Some deterioration in labeling was observed on aged fingerprints on glass and aluminum foil substrates.

Phloxine B, Nanocomposite, Fingerprint

D28 Blunt Cardiac Rupture Caused by a Rubber Bullet

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The goal of this presentation is to raise awareness about the fatal risk of a rubber bullet through the case of a Chinese fisherman's death in Korea. A variety of rubber bullets have been used in the world, and they have occasionally produced severe or lethal injuries. The shape and material of the bullets have been continually modified to minimize fatalities; however, it also can cause severe injury.

This presentation will impact the forensic science community by strengthening familiarity with rubber bullets and the injuries they cause with the increasing use of less-lethal weapons. It can also help the users of rubber bullets understand the nature of the weapon.

Rubber bullets are less-lethal weapons which are used to incapacitate dangerous people. The term "rubber bullets" commonly describes impact munitions made of rubber or other elastic materials, and they are one of the oldest less-lethal weapon technologies using kinetic energy. Despite the fact that it is designed to be safer than live ammunition, several cases of fatalities have been reported. Most of these fatalities happened because of abuse of the weapon in terms of range of fire and anatomical area of the body targeted. The 40mm bullet made of sponge foam, which is considered as an improved type of rubber bullet, is not likely to cause penetrating injuries because of the large size of the elastic round nose. But it can also cause severe internal injury. This is a case report of such a fatality following a rubber bullet injury. The Korean coast guard pursued an illegal Chinese fishing boat in South Korea's Exclusive Economy Zone (EEZ). The Chinese crewmen wielded various lethal weapons including saws, knives, and other items in their struggle with the squad members. With their lives threatened, the coast guard fired rubber bullets toward crewmen; the distance between the officer and crewmen at the time of firing was eight to ten meters. One crewman was struck on the chest and collapsed on the deck. The crewman who had fought for over ten minutes suddenly relented. He was transferred to the hospital, but died shortly after admission. The bullet was an improved projectile, which was 60g in weight and consisted of a sponge foam nose that was 40mm in diameter and had a plastic body. Through autopsy, contusions and several abrasions in the lower left central region of the chest were found. He had rib fractures and a 2mm rupture at the apex of the heart. The injuries of the heart and anterior chest were consistent with injuries caused by the rubber bullet. There were no signs of blunt force trauma from assault and he did not have any chronic diseases.

Rubber Bullet, Cardiac Rupture, Autopsy

D29 An Epidemiological Study of Medicolegal Organophosphorus Poisoning in the Central Region of Nepal

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After attending this presentation, attendees will gain an understanding about organophosphorus poisoning, its impact, prevention, and careful handling.

This presentation will impact the forensic science community by providing results from a two-year retrospective study in the central region of Nepal with very little previous research. This presentation will add to research being carried out in forensic toxicology by broadening the understanding of how to control the spread of organophosphorus pesticide and the proper use in crop protection.

Organophosphorus (OP) pesticide self-poisoning is an important clinical problem in rural regions of the developing world and kills an estimated 200,000 people every year. Acute poisoning by OP compounds is a major global clinical problem, with thousands of deaths occurring every year in Nepal. Methyl parathion (Metacid) and Dichlorovos (Nuvan, Doom) are commonly ingested OP pesticides; Malathion, Dimethoate (Rogor), Phorate, and Monocrotophos are other less-frequently ingested compounds in Nepal.

OP compounds are chemical compounds containing carbon-phosphorus bonds, primarily used in pest control and are often persistent organic pollutants. They are easily accessible, thus they are commonly associated with suicide and accidental poisoning. In the present study, 171 patients with severe OP poisoning were admitted to the emergency ward of National Medical College Teaching Hospital and Narayani Sub-Regional Hospital, Birgunj, Nepal, from January 2010 to December 2011, and were studied for a detailed epidemiological and medicolegal analysis.

The mean age of the patients was about 28 years of age. Most of the admitted cases were suicidal as well as accidental in nature, women were the main victims, followed by children. Suicidal deaths due to ingestion of OP compound are very common in Nepal, especially in women. The reason may be the increasing stress in the family and economic constraints. Accidental deaths due to occupational exposure or inhalation of OP compounds are reported but, in these cases, the mortality rate is less than that of suicidal poisoning. Further study is needed by the government and national and international Non-governmental Organizations (NGOs) to evaluate this issue.

Organophosphorus, Acute Poisoning, Epidemiology

D30 Comparison of XRF, SEM-EDS, ICP-MS, and LA-ICP-MS in Forensic Glass Fragment Analysis

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After attending this presentation, attendees will learn the strengths and weaknesses of the techniques adopted in differentiating glass samples and the compounds which are useful in differentiation of glass.

This presentation will impact the forensic science community by the variety of techniques used to analyze glass fragments.

Glass is one of the most tangible pieces of evidence found in forensic cases, as it appears everywhere in our daily lives, and, because of its physical, chemical, mechanical, and optical properties, it presents the kind of evidence that can be analyzed using many different methods. For this reason, much research has been conducted on these methods. This research has generally consisted of the determination of the glass refractive index and elemental analysis. Improvements in analytical technology have made possible the development of new methods in elemental analysis leading to an increase in the ability to distinguish between glass fragments. Because of its nature, glass consists of different oxides. When elemental analysis methods are applied, the oxide components in the glass vary proportionately. While some oxides make up a large bulk of the composition, others are found in very low quantities. The oxide composition of the glass is a very important factor in comparing different samples in terms of their relative oxide quantitative distribution.

The goal of this study is to establish the definition and differentiation of glass samples by looking at their elemental composition, particularly when the evidential glass is of unknown origin. In this study, glass samples encountered in many criminal cases are examined using X-Ray Fluorescence (XRF) spectrometer, Scanning Electron Microscope with Energy Dispersive X-ray Spectroscopy (SEM-EDS), Inductively Coupled Plasma Mass Spectrometry (ICP/MS), and Laser Ablation Inductively Coupled Plasma Mass Spectrometry (LA-ICP/MS). These four techniques provide high resolution of the elemental analysis of glass materials, while based on different principles.

A total of 50 uncolored glass samples were divided into two groups, window and headlight. As a result of some analysis that had been done during the research, it became clear that the quantification of only one oxide is insufficient to differentiate between the various types of glass. When several of the oxides are analyzed together, the results obtained can help in the recognition of and differentiation between glass fragments. Also, more precise results can enable determination of which oxide is useful in defining and differentiating the glass. This can be achieved by normalizing the elemental analysis of the oxide composition, selecting an appropriate oxide, and then carrying out a critical evaluation of the results.

In conclusion, this study will be performed in order to ascertain whether spectroscopic analytical methods are beneficial in distinguishing between glass samples and, thus, in defining the origin of unclassified glass.

Glass Comparisons, Elemental Analysis, Trace Evidence

D31 A Multiplex mRNA-Profilng System for the Forensic Identification of Body Fluids

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After attending this presentation, attendees will gain new insight about the identification of the biological samples that are often found at crime scenes by using mRNA analysis.

This presentation will impact the forensic science community by revealing that mRNA profiling is a promising method for the identification of various body fluids from biological stains with only one multiplex PCR reaction.

Forensic DNA analysis allows the identification of individuals from low amounts of stains containing only a few nucleated cells but DNA profiles have limitations on the determination of the type and origin of the evidence.

In many situations, this question could reveal the importance of establishing a link between evidence and the crime. In forensic practice, biological stains are routinely pre-analyzed with serological tests. Conventional serology-based methods for body fluid identification has many disadvantages, such as sample consumption, intensive exertion, time consumption, varying degrees of sensitivity and specificity, and no decisive tests for the presence of menstrual blood and vaginal secretions. Within forensic genetics, messenger Ribonucleic Acid Molecules (mRNAs) have increasingly gained popularity regarding their potential to distinguish human body fluids. mRNA profiling is a novel technique that has been investigated as a potential tool for human body fluid identification. Advantages of mRNA profiling include: the identification of a broad spectrum of body fluids; a single molecular-based method for virtually all body fluids; and the spontaneous detection of several stains by a multiplex Polymerase Chain Reaction (PCR).

This study describes the development of a single multiplex mRNA-based system for the discrimination of the most common forensic body fluids. As a result of this study, five different (blood, menstrual blood, semen, vaginal secretions, saliva) detections and typings of body fluid was intended. Therefore, hemoglobin beta (HBB) and b-spectrin (SPTB) for the blood, histate 3 (HTN3) and Stathern (STATH) for saliva, Prostate Specific Antigen/Kallikrein 3 (PSA/KLK3) and Semenogelin 1 (SEMG1) for semen, Matrix metalloproteinase 7 (MMP7) and Matrix metalloproteinase11 (MMP11) for menstrual blood, Humanbeta-to defensin1 (HBD1) and mucin 4 (MUC4) for vaginal secretions, gliseraldehyde-3-phosphate dehydrogenase (GAPDH) and beta 2 microglobulin (B2M) for housekeeping genes mRNA biomarkers were employed for differentiation of the mentioned body fluids.

Body fluids and tissues from ten individuals (five men, five women) were collected with their informed consent. RNA extraction was performed using the Dynabeads[®] mRNA DIRECT Kit and PureLink[®] RNA Mini Kit. RNA concentrations were measured using Qubit[®] RNA Assay Kit with the Qubit[®] 2.0 Fluorometer. The designation of primer sets were performed with Primer 3[®] software due to previously published references. The forward primers were 5'-labeled with 6-FAM, VIC, PET, and NED. Reverse transcription endpoint PCR used SuperScript[®] III One-Step RT-PCR System with Platinum[®]Taq on GeneAmp[®] PCR System 9700. PCR products were analyzed by capillary electrophoresis on a laser-induced fluorescence ABI[®] Prism 3130 Genetic Analyzer.

Successful identification and the discrimination of the body fluids were predicted on the experimental set-up. The

main objective of this study was to evaluate 12 mRNA markers by capillary electrophoresis for their specific identification of five human body fluid stains for forensic purposes. mRNA detection has been demonstrated to be a reliable method for positive identification of the most common biological materials obtained from forensic cases.

Forensic Genetics, mRNA Analysis, Biological Samples

D32 A Study to Determine the Use of Gunshot Residue Upon Clothing as an Item of Evidence

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After attending this presentation, attendees will gain an insight into the need to gain a primary level of experimental data for different types of trace evidence pertinent to an investigation in order to establish a contextual understanding of the behavior and persistence of such evidence. This approach then enables the incorporation of the results derived to build upon a secondary level experimentation on a case-by-case basis so as to provide an empirical base for the collection, analysis, and interpretation of trace evidence for intelligence and evidentiary purposes.

This presentation will impact the forensic science community by providing insights into the behavior of one particular form of trace evidence, Gunshot Residue (GSR), using Scanning Electron Microscope (SEM) analysis to identify the transfer, distribution, and persistence of GSR upon clothing.

In recent years, there has been a decrease in the use of trace evidence in criminal investigations. This is partly due to the discovery of DNA evidence and partly due to the lack of understanding into the behavior of the various types of trace evidence such as GSR. In order to gain such information, the aim of this United Kingdom-based study was to determine the behavior of GSR by analyzing its detectability after washing, by analyzing its spatial distribution upon clothing, and by determining a timeframe within which detectability is still possible. These are interesting factors to analyze as there is currently a limited amount of information that is inferred from GSR evidence; therefore, further understanding is needed to increase its evidentiary value and to be used as an evidentiary tool in criminal investigations.

This experimental study was designed to assess both spatial and temporal aspects of GSR behavior on clothing. First, clothing of three witnesses in close proximity to a shooter at different angles was analyzed. The firearm, a .357 Magnum[®] revolver, was discharged five times for each of the four experiments. Sampling was carried out by utilizing SEM stubs to tape-lift the GSR particles from various areas of the clothing. After sampling, each stub was analyzed by the SEM to confirm the presence of GSR.

Second, it was determined that after washing the clothing, the concentration of GSR was significantly reduced, making detectability unlikely but not impossible. It was also determined that there was a pattern to its spatial redistribution depending on the location of the witness. However, this distribution is dependent on a variety of variables that need to be considered if a secondary level of experimentation were to occur. In this study, GSR was still detected on the clothing after 12 hours. The implications of this finding, given that current regulations in the United Kingdom are that garments are only tested for GSR up to six hours after an incident, will be discussed.

This research, therefore, offers a positive foundation for further studies in order to determine a framework for GSR behavior

and its utility as a form of evidence in criminal investigations. It also shows that the variability in the GSR distribution pattern regarding garments should not be overlooked when recovering evidence.

Gunshot Residue, Trace, Forensic Evidence

D33 The Effects of Chemicals Used in Fingerprint Development Techniques on DNA Analysis

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After attending this presentation, attendees will be informed about DNA analysis contaminated with bloody fingerprints.

This presentation will impact the forensic science community by providing information to crime scene investigators and others concerning their access to a wide variety of chemicals for the enhancement of latent fingerprints in blood.

Mixtures composed of several biological samples are frequently encountered at crime scenes. Bloody fingerprints play an especially important role in forensic individualization and identification. Both blood and latent prints could reveal the interaction between perpetrator and the crime scene since they are unique and specific. Blood-contaminated fingerprints were assumed to be a challenging situation by crime scene investigators and analysts because it is difficult to obtain both items of evidence together during crime scene investigations since biological samples have different expiration periods and different methods are used on different samples that might affect the chemical structure of the biological samples. Eventually, the chemical reagents used in the development of fingerprints were thought to influence the extraction and analysis of DNA obtained from blood samples. Fingerprint detection and analysis for individual identification have undergone tremendous changes since their introduction to the investigation of crimes in 1892. Growing technology revealed an increase of several laboratory techniques. However, precautions and great care must be taken during investigating fingerprint evidence contaminated with any body fluid collected from a crime scene. Thus, the reagents used in developing the contaminated fingerprints play an important role for the degradation of the body fluids. The main goals of the study are to evaluate the integrity of the biological samples when exposed to reagents employed in fingerprint development and to investigate the effects of the mentioned reagents on DNA analysis. This study intended to determine the effect of 11 blood enhancement reagents on the subsequent Short Tandem Repeat (STR) DNA analysis of aged bloody fingerprints deposited on metal surfaces. Amido black, Rhodamine-6G, 1,8-diazafloren-9-one (DFO), Indandione, Ardrex™, 5-Methylthioninhydrin (5-MTN), cyanoacrylate, Thermanin, zinc chloride, silver nitrate, and ninhydrin were tested on galvanized metal plates with a series of depletion. DNA samples were extracted using the GF-1® Forensic DNA Extraction Kit. Samples were amplified by using the AmpF!STR® MiniFiler™ PCR Amplification Kit on thermal cycler and

were analyzed with ABI 3130 capillary electrophoresis. According to the results, while insufficient amounts of DNA were extracted from specimens treated with DFO, ninhydrine Rhodamine-6G, and zinc chloride, competent amounts of DNA for STR analysis were extracted from the specimens that were exposed to remaining fingerprint development methods. This situation can be explained by the damage of the chemical reagents targeting the biochemical structure of the DNA molecule. The development methods that acquired the highest amounts of DNA extract were Thermanin (1.13ng/µl) and cyanoacrylate (0.625ng/µl), respectively. Crime scene investigators now have access to a wide variety of chemicals for the enhancement of latent fingerprints in blood. As DNA typing technologies became more sophisticated and more sensitive, much smaller blood marks have been submitted for genetic analysis. This study revealed that some loss of biological material will take place with enhancement, specifically with procedures that require destaining steps, such as Rhodamine-6G, DFO, ninhydrine, and zinc chloride. The quality of fingerprints also depends on the method used. Several methods were used in the experiment. The results showed that the most effective method for developing fingerprints on metal objects was Thermanin.

DNA Analysis, Fingermarks, Blood

D34 Analysis of Cyanoacrylate Fumigation's Effects on the FTIR Classification and Comparison of Polymers

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After attending this presentation, attendees will understand the effects fumed cyanoacrylate has on polymer samples and how that impacts the infrared spectral data used for comparisons and classifications.

This presentation will impact the forensic science community by explaining that cyanoacrylate is a polymer and can potentially react with other polymers such as paint chips or synthetic fibers found in trace evidence via crosslinking or electrostatic attraction. The micro-Fourier Transform Infrared (FTIR) spectral data found affects the process of determining classifications and comparisons of trace evidence.

Cyanoacrylate fumigation is a common method used in forensic science to develop and preserve latent fingerprints on evidence either in the field or at the laboratory. Three items are necessary to perform this method: a viable substrate of cyanoacrylate such as commercially purchased superglue; a heat source to vaporize the cyanoacrylate substrate; and an enclosed system to provide an environment with the proper humidity. This process is commonly used in the forensic science field because these materials are affordable and can be easily used at crime scenes. Cyanoacrylate fumigation reacts with the amino acids found in sebaceous secretions of the fingerprint and is one of the first methods performed on evidence because of its ability to both develop and preserve latent prints.

The samples analyzed in this research study were prepared uniformly to closely mimic trace forensic laboratory evidence. This procedure minimizes the effect sample size may have on the fuming process and FTIR spectral analysis. Eighteen random paint samples were chosen and shaped under a stereoscope using a scalpel. The samples in this study were weighed, and an average mass was determined to allow sampled polymers from different sources to be uniformly prepared.

A comparison study between fumed and non-fumed

polymer samples was performed to evaluate the effect cyanoacrylate fumigation has on the FTIR spectral data and analysis. Polymer samples were collected from six different sources: tool paint; spray paint; architectural paint; automobile paint; glitter; and synthetic fibers. A triplicate of each sample was placed on a separate microscope slide and fumed with ASI® Adhesive Systems RP100 commercial superglue for the same duration of time. Non-fumed controls for each polymer along with a control sample of isolated fumed cyanoacrylate were collected. All samples were analyzed using a Thermo Nicolet® Continuum FTIR microscope instrument. The fumed polymer data was compared to the non-fumed control data, and conclusions were determined based upon the addition or absence of cyanoacrylate peaks.

Future research can be done in developing a non-destructive clean-up method to remove the cyanoacrylate from trace polymer samples. This would allow latent fingerprints to be developed and preserved first without impacting the following analysis of trace evidence. Another study could determine the penetration ability of cyanoacrylate on multi-layer paint, i.e., automobile paint. A clarification on whether cyanoacrylate infrared peaks can be seen in more than just the top layer spectra is needed.

The opinions or assertions contained herein are the private views of the author and area not to be construed as official or as reflecting the views of the Department of the Army or the Department of Defense.

Cyanoacrylate Fumigation, FTIR, Polymer Classification

D35 The Recovery of DNA in Indoor and Outdoor Area Environments

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After attending this presentation, attendees will have an understanding of how scientific techniques such as Polymerase Chain Reaction (PCR) are used in detecting the presence of pig (*Sus scrofa*) mitochondrial DNA collected from intestines of maggots.

This presentation will impact the forensic science community by identifying how environmental factors can influence postmortem changes and affect the presence of DNA on remains. In order to further explore the effects of indoor and outdoor areas, the decomposition and insect colonization of pig carcasses will be observed over a 42-day period in a semi-arid environment located at Research Site located in Lubbock, TX.

The focus of the study is to analyze samples collected from the intestines of early in-star maggots used to identify the presence of pig (*Sus scrofa*) DNA in decomposed remains placed in an open-air environment and in an enclosed environment. This study hypothesizes that the amount of DNA recovered will vary significantly between the two environments.

This study involves two female pigs that are being used as an animal model for human victims, suspects, or third-party suspects for forensic investigations. Deoxyribonucleic acid sequence data will be obtained from the intestines of maggots that feed on pig remains. Deoxyribonucleic acid has become standard in forensic science to analyze biological samples from decomposed remains. In addition, DNA serves as an essential tool for the identification of humans.

Two juvenile female pig carcasses weighing approximately 15kg will be placed at Research Site; one will be placed inside a wooden house and the other will be placed outdoors in a field. Observations and sample collection from the pig carcasses will be conducted daily. Blowfly (*Calliphoridae*) larvae (maggots)

will be collected in late summer and the intestines of the insects will be analyzed to determine the amount of DNA present. The characteristics of a semi-arid environment are such that developmental growth of maggots may be impeded, consequently having a possible influence on the amount of DNA recovered. Maggots will be collected and preserved in 15ml falcon tubes filled with 5-10ml of 70% ethanol. All larvae samples collected will be placed in a freezer at 4°C to improve preservation of the DNA in maggots and stored until ready for extraction.

Following DNA extraction, a PCR assay will be used to identify the presence of DNA recovered from the maggot's intestines. This procedure identifies and purifies the DNA recovered from samples and can be used as evidence to solve criminal cases such as homicide, sexual assault, and negligence. Gene sequences will be amplified by PCR, sequenced, and analyzed by capillary electrophoresis. The expected findings of this study may determine that arid environments can affect the amount and recovery of DNA. In addition, the amount of DNA recovered is expected to be higher in an outdoor environment versus indoor environment. Therefore, these results can determine that environmental factors may show differences in decomposition patterns that can influence the recovery of DNA in remains.

Arid Environment, Decomposition, Mitochondrial DNA

D36 Who Was Holding the Gun When the Fatal Shot Was Fired? Deductive Application of Crime Scene Work

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After attending this presentation, attendees will better understand the value of experimentation and collaboration of disciplines to prove or disprove a theory involving criminal investigations. Viewers will be presented information about a specific case where a young adult female was shot at close range with a handgun and it was disputed if she or her boyfriend held the firearm when it was fired. Determining the minimum and maximum range of the muzzle to the wound, coupled with deductive reasoning, were key to proving the manner of death.

This presentation will impact the forensic science community by highlighting the fact that the deductive application of experiment findings, facts, and crime scene work is required in order to find the truth.

On September 4, 2010, a young adult male called the police stating his girlfriend had shot herself in his apartment. First responders found the female deceased on the living room carpet and a handgun nearby on a table. The male claimed his girlfriend and he had been drinking heavily, and when she handed the handgun to him, it "went off" and she dropped to the ground. He was unclear who was holding it at the time of discharge. He claimed they were facing one another, standing in the living room, and the shooting was an accident. The scene investigation uncovered a secondary impact strike by the projectile at 41 inches in height, indicating a downward trajectory after it passed through the victim. The recovered semi-automatic Taurus® .40 caliber handgun had a cartridge chambered with a magazine loaded at three rounds less than capacity. One casing and two cartridges were found to the right of the male, consistent with their having been ejected while the firearm was in the same orientation as it was at the time of its fatal discharge. The autopsy showed a downward trajectory through the victim's right eye area, exiting the center back of her head, and the presence of stippling. Subsequent distance determination placed the muzzle of the firearm between three and 15 inches at

time of discharge, but most likely between six and 12 inches. The investigation revealed a previously unreported incident of the male threatening the female with an unloaded gun and pulling the trigger.

The manner of death is key to every death investigation and is a crucial part of legal proceedings, both criminal and civil. Determining the manner of death is not always easy, and when a suspect's statement is vague, the outcome of an investigation is in jeopardy due to a multitude of possible manners of death. Using science to prove a minimum and maximum range, the fact that the muzzle of the gun could have been from the victim was only part of the puzzle. Measurements of the suspect's and the victim's arms, their height, and a secondary impact point of the projectile helped narrow the range of possibilities. The location of both the spent casing and the firearm and the condition of both participants' hands proved to be the key determining factors. A scene recreation with photos and diagrams to show the range of possibilities made it easier to understand and "see" the truth.

In the legal arena, the exclusion of possibilities can be as important as the inclusion of them, and inductive logic is not desired. The use of experimentation and deductive logic allowed the decedent to receive justice.

Theory, Experimentation, Deductive Logic

D37 Multi-Element and Stable Isotope Analyses in Hair

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After attending this presentation, attendees will gain valuable Korean data that they can compare to their own data. They can also discuss how to obtain information concerning occupations, sex, and smoking habits from the data.

This presentation will impact the forensic science community by providing data from Korean people because a variety of data from many countries is needed for identification.

The application of Isotope Ratio Mass Spectrometry (IRMS) and Laser Ablation Inductively Coupled Plasma Mass Spectrometry (LA-ICP/MS) to forensic hair samples was investigated. Identification of forensic hair samples was carried out using stable isotopes and multi elements, as well as examining what types of information can be developed from hair samples using these analyses. Isotopic data (C, N, O, S, and H) for bulk hair indicated that this signature can be used as an alternative tool of identification in cases where no material is available for DNA comparison or where no DNA match can be found. The single hair strand was analyzed for information such as sex, smoking habits, and occupation using LA-ICP/MS. Even though this study was preliminary, good results were obtained.

Hair specimens from 16 people (5 females and 11 males) living in Seoul and the Gyunggi province were collected for this study. The scalp hair samples were stored in glass vials at room temperature before washing. The results are as follows: (1) gender can be determined by the amount of Hg and Pb in hair; (2) smokers are distinguished from non-smokers as the content of La and Na in the hair of smokers is higher than non-smokers; and, (3) the samples are divided into four groups depending on occupation. The amount of Al is high in people working at the laboratory and the amount of Hg and Na is high in fishermen. The content of Sn is high in painters and the content of Mn and Fe is high in blacksmith

workers. These results indicate that the concentrations of metals in hair are influenced by the environment of an individual work place; and, (4) bulk stable isotope analysis of hair can provide information concerning a person's dietary intake and the geo-location in which a person has lived. These isotope results will have forensic application. The average of $\delta^2\text{H}(\text{‰})$, $\delta^{18}\text{O}(\text{‰})$, $\delta^{13}\text{C}(\text{‰})$, $\delta^{15}\text{N}(\text{‰})$, and $\delta^{34}\text{S}(\text{‰})$ is -75.6 ± 5.3 , 11.8 ± 1.1 , -19.1 ± 0.4 , 8.8 ± 0.6 , 9.8 ± 0.5 , respectively. These results indicate that Koreans have their own chemical fingerprint even in hair.

Further study with more hair samples is needed. In cases using the LA-ICP/MS standard, fewer methods (including Aridus) or house-working standard methods should be examined.

Multi-Element, Stable Isotope, Hair

D38 Population Genetic Data of 17 Y-STR Markers in Turkish Cypriots From Cyprus

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After attending this presentation, attendees will learn more about recent Y-chromosome analyses conducted on Turkish Cypriots from the island of Cyprus and see how this new Y-STR haplotype dataset may also contribute to a better understanding of the population genetics of the Eastern Mediterranean Basin.

This presentation will impact the forensic science community and the field of population genetics through the introduction of population data on 17 Y-STR loci for 206 individuals from the Turkish Cypriot population, which constitute the largest dataset reported from Cyprus so far and also from a region that is still relatively understudied.

Cyprus is the third largest island in the Mediterranean Sea situated 75km south of Turkey, 108km west of Lebanon, 380km north of Egypt, and 800km southeast of the Greek mainland. Today, there are two major ethnic groups on the island, namely the Turkish Cypriots who speak Turkish and are Muslims and the Greek Cypriots who speak Greek and are mostly Orthodox Christians. The subject of this study, the Turkish Cypriot population, resided all over the island in villages or cities consisting of either mixed ethnic background or exclusively Turkish Cypriot up until 1963-1974, but then largely relocated to/concentrated in the Turkish Cypriot-administrated North Cyprus since 1974. This study has analyzed 17 Y-chromosomal Short Tandem Repeat (STR) loci included in the AmpFISTR® Y-filer™ system (DYS456, DYS389I, DYS390, DYS389II, DYS458, DYS19, DYS385a/b, DYS393, DYS391, DYS439, DYS635, DYS392, Y-GATA-H4, DYS437, DYS438, and DYS448) in 206 unrelated, healthy male individuals from the Turkish Cypriot population currently residing in North Cyprus. Among the 206 haplotypes observed, there are 198 unique haplotypes and 8 haplotypes that were found in two individuals each. While no locus duplications and null alleles were detected, 39 allelic variants in total were observed, the majority of which (22 out of 206 haplotypes or 10.68%) were comprised of intermediate variants at the DYS458 locus (alleles 16.2, 17.2, 18.2, 19.2, and 20.2). For the 198 unique haplotypes observed in the Turkish Cypriot dataset, a Discrimination Capacity (DC) of 0.9611 was observed, with an overall Haplotype Diversity (HD) of 0.9181, whereby the calculations included DYS389I and DYS389II allelic values as is and DYS385a/b alleles as diploid values. The calculated average Gene Diversity (GD) values ranged from 0.3576 to 0.9622 for the DYS392 and DYS385a/b loci, respectively. A comparison of the Turkish Cypriot Y-STR dataset with those already

available from historically and/or geographically related countries (e.g., Turkey, Lebanon, Egypt, Greece, and Italy) through the use of Analysis of Molecular Variance (AMOVA) confirmed that this data does not deviate significantly from the typical core haplotypes of the region. While the Turkish Cypriot Y-STR haplotype dataset will find immediate use in the Committee on Missing Persons in Cyprus Project on the "Exhumation, Identification and Return of Remains of Missing Persons," it is also expected to contribute to the establishment of forensic genetic services in North Cyprus and to be useful for the global forensic genetics community in general.

Population Data, Y-STRs, Turkish Cypriots

D39 Estimation of Postmortem Interval Using Thanatochemistry and Postmortem Changes

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WITHDRAWN

D40 Andragogical CSI: Strengthening the Education and Mentorship Into the Future

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After attending this presentation, attendees will be provided with the constructs and development of a comprehensive training institute that takes crime scene practitioners from basic training modules, through more specialized training, and into more professional development opportunities that promote continued advancement throughout an individual's career. Most significantly, this presentation will contribute to the recommended changes to the field of crime scene investigators, which are outlined in the National Academy of Sciences (NAS) Report, *Strengthening Forensic Science in the United States: A Path Forward*.¹ The presentation details many additional areas within the fields of criminal justice and forensic science training that need to be addressed, studied, and corrected, such as cost-effective continuing education, mentorship, and certification options for crime scene investigators. Effective leadership is crucial toward successfully aligning current policy, education, and practices with the NAS recommendations.

This presentation will impact the forensic science community by highlighting the development of a cost-effective continuing education institute that can be duplicated in other regions of the country, which will regionally foster growth of Crime Scene Investigators (CSIs) throughout their career, while also offering mentorship opportunities, areas of specialization, and professional growth into the future.

The Northeast Crime Scene Institute (NCSI) is a comprehensive continuing education program hosted by the Somerset County Prosecutor's Office Forensic Laboratory in Hillsborough, New Jersey. Created in October of 2011, the institute establishes its duties as providing crime scene investigation, analysis and forensic training programs for criminal justice personnel, and providing technical assistance to criminal justice agencies and aiding them in the discharge of their responsibilities. The institute offers three distinct modules that coincide with practitioner experience and development.

The first module consists of the Crime Scene Investigation

course. Drawing together the many fields of study in the forensic sciences, this course provides the standardization of basic skills and practices needed by crime scene investigators to successfully fulfill their duties and responsibilities. The course is provided annually, four weeks in length, with each week spaced intermittently throughout the year. The student's skills and abilities are assessed with lab performance evaluations, knowledge assessments (tests), and work experiences while establishing a mentorship with instructors that lasts approximately eight months.

The second module consists of various specialty courses that are designed to advance upon the basic skills and abilities of investigators gained from the first module. The course offerings at this level are geared to hone those abilities toward a content-specific field of analysis. A variety of courses that will be offered may include, but is not limited to, latent fingerprint analysis, bloodstain pattern analysis, medicolegal death investigation, and shooting reconstruction. These courses will normally consist of approximately one week of specialized training pre-scheduled annually.

The third module seeks to continue the advancement of practitioners toward professional contributions in the field of crime scene investigation. More specifically, the third module will assist interested participants in taking part in more advanced practices that will contribute to the advancement of instruction, research, publication, and administrative activities that are necessary for the NCSI to remain at the forefront of CSI continuing education. This module will offer courses and opportunities for members to hone their skills at instructing classes, publishing articles, conducting research experiments, and taking on voluntary roles within the administration of the NCSI.

Reference:

1. National Academy of Sciences. *Strengthening Forensic Science in the United States: A Path Forward*. The National Academies Press, 2009. Washington, D.C.
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Education, Training, Mentorship

D41 The Development of an Experimental Setup and Recovery of Biological Evidence From Bullets for DNA Analysis

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After attending this presentation, attendees will gain insight into the challenges that face DNA recovery from fired bullets.

This presentation will impact the forensic science community by providing a protocol for evaluating biological evidence on bullets.

The recovery of DNA profiles from firearm ammunition is of specific interest to law enforcement and the criminal justice system. Swabbing fired projectiles can be of high importance in the systematic and hypothesis-driven reconstruction of shooting scenes.

In incidents of mass shootings or when the crime scene scenario is in question, bullets bearing biological samples from "touch" or from penetrating wounds can become lodged into structures such as walls of various compositions, seat cushions, flooring, and other strata. DNA on fired bullets endures a severe environment of heat and friction, and inhibitors from swabbed bullet surfaces can have a negative impact on the amplification efficiency. Therefore, the recovery of DNA from bullet surfaces may be in question due to the quantity and the quality of recovered biological tissue. To address these challenges, the parameters that

can significantly impact the persistence and recovery of biological samples on fired bullet surfaces were considered. Priority was given to factors such as bullet composition and calibers, the nature of impact surfaces, and the effect of muzzle-to-target distance.

To initiate this long-term and multi-stage study, an experimental setup compatible with current human Short Tandem Repeat (STR) analysis platforms was devised. This setup consisted of biological tissue targets made of human blood (in ethylenediaminetetraacetic acid (EDTA)) and 2% agarose mixture at a ratio of 1:40 (blood to agarose). After cooling the agarose, the agarose-blood mixture was prepared and poured into a plastic container to form a 10.5cm-diameter target approximately 1cm in thickness. A portion of the 10cm x 12cm piece of sterile gauze was submerged in the mixture, leaving approximately a 10cm length of unsubmerged gauze. In ~10 min, the target set and adhered to the gauze inside the container. Then, using the extra length of gauze, the targets were removed from the container just before the bullets were fired. Next, one wall section was prepared using one piece of plasterboard and three pieces of dressed yellow pine. One piece of yellow pine was 3.8cm x 23.5cm x 2.44m and one piece of yellow pine was 1.9cm x 23.5cm x 2.44m. The plasterboard was 1.5cm x 23.5cm x 2.44m. The wall section was made by attaching the plasterboard to the pieces of yellow pine with screws and the plasterboard and the two yellow pine pieces were clamped together using C-clamps for transporting the wall section to the firing range. At the firing range, the biological targets were attached by stapling the extra length of gauze to the plasterboard side of the wall section. Handguns were used to fire .22 caliber, 9mm, and .38 caliber bullets into the targets at a distance of 1m. This distance prevented a significant portion of gunshot residue and gases from entering the target. The wall sections were disassembled and the bullets recovered for DNA analysis. Most of the .22 caliber bullets disintegrated with fragments <1mm in size. Consequently, there was insufficient surface area for swabbing from that caliber. The 9mm and .38 caliber bullets were collected and swabbed with COPAN® 4N6 FLOQSwabs™ in order to recover biological samples for DNA analysis. DNA samples were extracted using the COPAN® Nucleic Acids Optimizers (NAO), a semi-permeable basket, which retains fluid until centrifuged, with the PrepFiler Express™ and PrepFiler Express BTA™ Forensic DNA Extraction Kits. DNA quantitation was carried out using the Quantifiler® Human DNA Quantification Kit. The AmpFLSTR® Identifiler® Plus PCR Amplification Kit was used for Polymerase Chain Reaction (PCR) and the 3130 Genetic Analyzer™ was used for analysis. The quantitation results show that over 90% recovery of biological sample was achieved from the 4N6 FLOQSwabs™ swabs. While several swabs from bullets resulted in detectable DNA profiles, none of the profiles were attributable to the target with human blood. The results illustrate the utility of the experimental setup, reveal the challenges of biological evidence recovery from fired bullets, and pave the way for further testing.

DNA Evidence, Bullet Recovery, Bullet DNA

D42 Embracing Technology in Forensic Science Higher Education

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After attending this presentation, attendees will learn how technology and interactive online learning modules are integrated into the University of Ontario Institute of Technology (UOIT) Forensic Science Program undergraduate curriculum.

This presentation will impact the forensic science community by highlighting current trends in student learning.

The use of interactive digital content, tablet, and iPad® computer technology will be discussed.

In the UOIT Forensic Science program, a strong emphasis is made on developing students' practical skills. It is important for undergraduate forensic students to become well-versed in the fundamentals and in applying that knowledge. Hands-on, repetitive learning helps to reinforce these skills and concepts.

Combining online curriculum with traditional teaching styles promotes active learning. Interactive digital content is a tool for educating both inside and outside of the classroom. By focusing less on the fundamentals inside the laboratory, students can participate in a higher level of learning and critical thinking since the knowledge base has already been acquired. The advantages of digital content include: subject matter lends itself to repeated use and trial-and-error; students determine how they navigate through the learning object and develop the skills of sequencing and selecting tasks; students are allowed to move forward through the material at their own pace and review material as they deem necessary; and assessments are external to the learning object, permitting its use in a variety of courses of varying breadth and depth.

Technologies in terms of digital content and hardware have been embraced by UOIT's Forensic Science faculty. Several learning objects have been integrated into the academic curriculum and include, for example: (1) forensic document analysis; (2) digital photography; and, (3) crime scene processing. Also, both tablets and iPads® have become fundamental tools in both lectures and laboratories, having replaced pen and paper in the traditional sense.

An initial survey of undergraduate forensic students concludes that these technologies offer a valuable supplement to traditional teaching material and all agree in their merit as successful self-directed learning tools.

Forensic Education, Technology, Digital Content

D43 Honor Crimes: Definition, Diagnostic Criteria, and Risk Factors Through Presentation of Case Reports From Germany

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After attending this presentation, attendees will understand the meaning of the term "honor crimes" through the introduction of a scientific definition that has been proposed by a study. Causes, potential risk factors, and criteria of diagnosis will be explained through the discussion of three case reports that took place in Germany during the last 13 years.

This presentation will impact the forensic science community by introducing a scientific definition for honor crimes which explains their most important diagnostic criteria and potential risk factors. This will draw the attention of forensic examiners to such cases, hence saving potential victims and helping combat this phenomenon.

Honor crimes are one of the forms of domestic violence to which women are exposed in many countries. This type of crime

is difficult to diagnose because until now there were no specific diagnostic criteria to guide the forensic examiners in their diagnosis.

In this study, a scientific definition for honor crimes is proposed. Murders that were categorized as honor killings by German media in both Hamburg and Berlin in the last 13 years were retrospectively studied. The reports of the police, prosecution, and clinical forensic medicine as well as autopsy reports of 20 cases were examined. Among these, only one case in Hamburg and two cases in Berlin were identified as honor crimes according to the proposed definition.

The first case occurred in 2002 in Berlin when a 26-year-old Turkish female was stabbed and murdered by her 19-year-old brother. At the age of 14, the victim's family forced her to move from Germany to Turkey and marry a Turkish man. Two years later, the married couple moved to Berlin. They were then separated and the victim moved to a separate apartment. The victim then started dating a German man of whom her family didn't approve. Finally, her brother killed her in the apartment of her German boyfriend.

Three years later another Turkish female, 23-years-old, was shot in the street in Berlin by her 18-year-old brother. The victim's family had forced her to move to Turkey at the age of 15 to marry her Turkish cousin. She then separated from her husband, returned to Berlin, and took off her veil. She suffered from repeated violence and abuse from her family because she refused to observe their conservative traditions. She was moved to a women's shelter to protect her from her family, but this didn't stop her brother from killing her.

The third case took place in Hamburg in 2008, when a 16-year-old Afghani girl was stabbed and killed by her 23-year-old brother. The cousin helped the brother murder the victim, and afterward they left her in the street to die. The victim was repeatedly beaten and abused by her parents and her brothers because they didn't approve of the way she used to dress and because she had many male friends. She was moved to a women's shelter which she left several times to return to her family. During the trial, the brother willingly admitted killing his sister to cleanse the family's honor.

The three victims were killed by their brothers and, in two cases, the female victims were moved to women's shelters to protect them from their abusive families. Moreover, two cases were forced to get married at a very young age.

Domestic violence and forced marriage are types of violence that are being practiced against women in many parts of the world. In honor crimes, torturing and killing of the culpable female are legitimized by the family members and are considered as a necessity to cleanse the family's honor that has been besmirched by the female. In such cases, the victims have nobody to turn to for protection and eventually become victims of murder. Increasing attention to such crimes will allow the forensic community to deal with cases of domestic violence in more depth knowing that those victims could be potential victims of honor killing.

The presentation of these three cases will eventually be part of a larger study involving honor killings in all of Germany to reach common diagnostic criteria and risk factors that characterize these crimes.

Honor Crimes, Violence Against Women, Forced Marriage

D44 Child Abduction Murder: Time to Death by Victim Age-Group Category and Distance Between the Body Recovery Site and the Homes of the Victim and Offender

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After attending this presentation, attendees will gain an understanding of how quickly abducted children are murdered by age category and the proximity of an offender's home to the body recovery site and other murder-incident components (victim last seen site, initial contact site, and murder site).

This presentation will impact the forensic science community by adding information to an area with little prior research. Specifically, this presentation will identify locations where distance intervals may be more likely to produce relevant forensic evidence linking the offender to the victim, improving the effectiveness and efficiency of the investigations. This presentation will also give investigators a better idea of how long differing age categories of children are kept alive after they are abducted.

There are few empirical studies on child abduction murder.¹⁻⁴ Little research has addressed the influence of time and distance on case solvability in murder investigations of abducted children.^{1,2} Only one study has addressed the impact of forensic evidence on child abduction murder investigation solvability.⁵

The relationship of time and distance between child abduction victims and offenders was explored by examining child abduction murders occurring from 1968 to 2002 ($N=735$). Information from each case relating to time spans and intervals of distance between the offender's home and the victim's homes and other murder incident components were analyzed. In addition, the time between when the child was abducted and when they were killed was examined by victim age-group category.

Preliminary results indicate that as the victim's age increases, the time span between when the victim is abducted and when the murder occurs does not always increase. Abducted children who were murdered were killed within three hours (76.2%; 1-4). Young children between the ages of 0 to 5 years old were killed within three hours at a higher percentage than the other age groups (81.8%, $p > .05$). Young children (0 to 5 years old) were killed more quickly than middle childhood victims (6 to 11 years old), young teenagers (12 to 14 years old), and older teenagers (15 to 17 years old).

Previous solvability research has shown that the more investigators know about the distances between the murder incident components (victim last seen site, initial contact site, murder site, and body recovery site), the more case solvability increased.¹⁻⁴ However, these studies did not examine the distance between the victim's home, offender's home, and the body recovery site. Approximately 37.3% of the victims' bodies were recovered within one-and-one-half miles from the victim's home and 38.7% of the bodies were recovered within one-and-one-half miles from the offender's home. In addition, the study explored the distance between the victim's home and offender's home and other murder incident components. The findings provide valuable information to investigators in the absence of other leads or evidence.

Most child abduction murder victims are victims of opportunity; therefore, knowledge about the distance probability between the offender's home and the body recovery site, as well as the distance from the body recovery site and victim's home, may provide valuable investigative direction if no other leads are available. Findings suggest that area searches of at least one-and-one-half miles should be conducted during an abduction investigation. Results also indicated that further research should be conducted to explore how forensic evidence recovered from these additional murder incident locations (offender's home and victim's home) may impact solvability. This examination adds to the literature on how time and distance operate within murder investigations. Because time and distance are critical solvability factors in child abduction murder investigations, this study is a valuable investigative tool for homicide detectives.

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Child Abduction Murder, Time and Distance, Forensic Evidence

D45 Scenes Are Not What They Seem! A Case Study

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This presentation will education investigators on one's approach to crime scenes and provide a variety of questions that may sort out some confusion and outright false information provided during the early stages of the investigation.

This presentation will impact the forensic science community by providing the opportunity to look at what biases may be brought to a crime scene. The forensic science community, especially pathologists who take into consideration the scene photographs and preliminary circumstances surrounding the event, will appreciate the need for clarity and specificity regarding scenes in order to assist in autopsies. Also, attendees will be able to enhance their investigative knowledge and acquire alternatives to approaching a scene.

The world is changing and becoming more populated with different cultures, mores, and practices. Well, so are crime scenes. People do unimaginable acts on other people and living creatures. Society has become private voyeurs with the increase in crime scene shows and documentary-style programming, such as the ID channel's *Wives with Knives* and Oxygen® Channel's *Snapped*. Harris County, Texas, is but a small part of this world of death and destruction, despite the fact that the total number of deaths each year makes it appear Harris County is near the top nationally with overall deaths.

Harris County has over four million people and autopsies approximately 4,000 cases each year. In 2012, there were 4,041 medicolegal cases reported to the Harris County Institute of Forensic Sciences; investigators responded to 1,652 scenes. The types of scenes responded to included: residences; office buildings; roadways; vehicles; open fields; and inside containers. Most scenes are consistent with witness statements and/or law enforcement's preliminary investigative reports; however, many are not.

Harris County is a large jurisdiction with active commerce,

and crime scenes are becoming more graphic, hidden, and complicated. One aspect of death investigation that has been changing over time is that those scenes are not what they seem. This presentation will outline a scene investigation that started out as a possible sexual assault, breaking and entering, drug abuse, and domestic violence. This scene required trace collection and took a completely different and innocent turn as the autopsy was underway. Scene and autopsy photographs will accompany the presentation.

Crime Scene, Deceiving, Case Study

D46 Law Enforcement and the Missing/ Unidentified Person Crisis

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After attending this presentation, attendees will have a better understanding of the complexities of the missing and unidentified crisis in the United States and a protocol to manage this crisis.

This presentation will impact the forensic science community by giving the law enforcement community forensic tools to handle this never-ending mass fatality disaster.

If you ask most Americans about a mass disaster, they're likely to think of the 9/11 attacks on the World Trade Center, Hurricane Katrina, or the Southeast Asian tsunami. Very few people, including law enforcement officials, would think of the number of missing persons and unidentified human remains in our nation as a crisis. However, it is what experts call the nation's silent mass disaster; a mass fatality disaster over time and space.

Should law enforcement agencies be in the missing and unidentified person business? The missing and unidentified persons have, over the years, been thrust upon law enforcement from the largest to the smallest of agencies. Neither is equipped to handle what is essentially a forensic problem — identification of the unidentified via scientific evidence: DNA; fingerprints; medical and dental X-rays. Solving cold cases is not a matter of chance or luck; it is quite simply a matter of hard work, design, protocol, and the passion of the investigator. Law enforcement is neither equipped for nor do they want the job. Law enforcement agencies are understaffed, under budget constraints, under time limitations, and consider the missing person and unidentified as a very low priority. It is estimated that there are over 100,000 missing persons and approximately 40,000 unidentified deceased persons in the United States. The fact that no one actually "knows" the exact numbers is itself a condemnation of the current handling of the missing and unidentified cases in the United States.

Sir William Gladstone (1809-1898), the former Prime Minister of Britain, said, "Show me the manner in which a nation or community cares for its dead and I will measure with mathematical exactness the tender sympathies of its people, their respect for the laws of the land, and their loyalty to high ideals." Improvements can be made to the current system and the National Missing and Unidentified System (NamUs) is a step in the right direction. This presentation will describe the protocol used to process missing and unidentified persons cases by a medium-sized law enforcement agency and medical examiner's office over the past ten years. The presented protocol may serve as a "model" for investigating missing and unidentified person cases at a local/county level and provide attendees with the tools necessary to implement a similar system in their jurisdictions.

Missing, Unidentified, Protocol

D47 Providing Forensics Training and Mentorship to the International Community

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After attending this presentation, attendees will have a better understanding of some of the challenges, pitfalls, and potential successes of providing forensic science training/mentorship programs in less-than-traditional laboratory environments under sometimes extremely austere conditions.

This presentation will impact the forensic science community by detailing "lessons learned" from early forensic outreach efforts that began with support from U.S. and other government forensic agencies in military theaters of operation. These efforts went beyond traditional "government-to-government" services, providing private industry with the opportunity to rapidly respond to an emerging need for improved forensic and biometric technologies outside the United States and North America.

These training and mentoring programs provide instruction in the latest standard operating procedures, protocols, technologies, and methodologies as they are applied to the various forensic disciplines and can be tailored to the needs of the host nation and its societal and jurisprudential requirements. As rule of law is becoming more of a focus across the world, it is imperative to provide support to countries who are striving to establish standard practices and procedures. Programs of instruction/mentorship can also be geared toward direct training or discipline-specific "shepherding" of select host nation laboratory personnel depending on the wishes of the host government. Programs of instruction/mentorship are designed to meet the needs of the host country's existing forensic capabilities and not to burden those countries with limited resources and/or infrastructure. By providing them technologies and methodologies within their economic and educational limits, these programs provide a sustainable foundation for continued operational success.

As the host nation personnel become more proficient in the forensic techniques and analysis, the goal is for the instruction to evolve into a train-the-trainer scenario so they can continue to educate their own colleagues. The goal is for each nation to maintain their own laboratories without the assistance of an outside entity. Recent successful forensic and biometric training efforts in the Middle East and South America will be highlighted as well as ongoing outreach efforts in Africa and other non-traditional forensic support regions. As there is an effort within the international law enforcement community to strive toward the highest levels of professionalism and precision in criminal investigation, it is incumbent upon the forensic science disciplines to provide assistance when possible. Given the limited resources available in the realm of "government-to-government" service, the recent rise in private-sector forensic service providers is a viable alternative to provide these training/mentorship programs. Designing these programs to be economical, practical, and interactive with the host nation participants has led to increased awareness and demand for such services.

Forensics, Training, Mentorship

D48 Forensic Awareness for Emergency Department Staff

Jennifer R. Schindell, BSN, 1115 Jackson Street, SE, Albany, OR 97322*

After attending this presentation, attendees will understand key concepts used to successfully teach emergency department staff the significance of their observations, documentation, and actions during the care of patients under conditions of potential forensic significance.

This presentation will impact the forensic science community by offering a sample curriculum for use in future trainings of emergency department staff.

Physicians, nurses, and technicians working in an Emergency Department (ED) commonly participate in situations of forensic significance and their contributions have the potential to dramatically alter the course of an investigation. The goal of this training is to elevate participants from a level of unconscious participation/obstruction to at least a level of basic forensic awareness in commonly encountered situations. The actions and observations of ED personnel can be particularly important in cases involving traumatic injury, neglect, overdose, Apparent Life-Threatening Events (ALTEs) in children, and any type of death investigation.

Though patient care must never be compromised in an effort to protect evidence, medical staff can be successfully trained to maintain a level of forensic awareness while providing care. The first step toward improving forensic awareness in the ED setting is to make clear the value of initial observations/ documentation and the potential ramifications of alteration to the body and related items. Information gathered during initial interactions can be of great value, not only to any subsequent investigation, but also to the provision of medical care. Because medical professionals receive little education regarding the pivotal role they may play in cases of medicolegal significance, this type of in-service education may be essential to the ED as a whole. A more thorough and accurate understanding of the circumstances surrounding a patient's arrival at the ED can only improve patient outcomes. In addition, providing this type of training decreases the likelihood that information will be lost or destroyed due to a mere lack of knowledge.

For this training to be effective, ED staff must receive information they interpret as relevant and realistic. Instructors of this curriculum should have experience in both ED and medicolegal settings. ED experience provides the instructor with an appreciation for structural barriers, as perceived by medical staff, and fluency in the "medical shorthand" care providers generally use to communicate. Each experience increases the likelihood of teacher-student dialogue and advanced problem-solving. Case reviews are an essential component of the medicolegal portion of this training and will allow attendees to better appreciate and personalize the information being given.

Improving forensic awareness in ED staff will not only improve efficiency and accuracy of subsequent forensic investigation, but also enhance patient care. While the primary obligation of the medical staff is to the health and safety of their patients, the patient and greater community are better served when forensic awareness is maintained. This presentation will review cases in which ED staff both aided and hindered forensic investigations. Examples of improved outcomes following this training approach will also be offered.

Evidence, Medical, Training

D49 A Multidisciplinary Data Fusion-Based Approach to Remote Human Grave Detection

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After attending this presentation, attendees will appreciate the use of data fusion for remote grave detection, and will be introduced to different types of data that forensic investigators might consider when searching for clandestine burials. This presentation will demonstrate the importance of maximizing data collection opportunities for achieving both evidence- and victim identification-based goals.

This presentation will impact the forensic science community by illustrating the growing role of imagery analysis in forensic research; in particular, the characterization of unique physical grave signatures to be used in future predictive modeling studies as well as anomaly or change detection.

Social conflict poses an urgent humanitarian threat to civilians that the international human rights community would like addressed. Globally, millions of missing people are thought to be buried in clandestine graves in both active and post-conflict zones throughout the world. Having knowledge of the locations of these mass graves may inform decisions to apply political pressure to offending parties, to plan interventions to impede or even halt regional human rights violations, or to assist in reconciliation efforts. Current research involving the University of Tennessee's Anthropological Research Facility (ARF) and the Geographic Information Science and Technology Group (GIST) at Oak Ridge National Laboratory is geared toward the development of a successful approach to the remote detection of graves so investigators are better informed during the management of past and present human rights violations.

The remote detection of human graves is largely under-studied, yet many air- and space-based imaging sensors demonstrate impressive performance when used to predict, and in many cases detect, anthropogenic ground disturbance, particularly in archaeology.^{1,3,4} Animal burial detection has been explored in the past by Kalacska and colleagues, who have demonstrated the potential benefit of using high-resolution spectral imagery in forensic contexts.² However, few other remotely sensed datasets have been tested in forensic research. Kvamme and colleagues have urged the archaeological community to combine, or fuse, data to increase the spatial, spectral, and temporal resolutions of remote sensing datasets — offering a logical argument that applies to many scientific disciplines — that data compiled from a variety of sensors and sources will paint a more complete picture of site composition and context, where physical qualities are otherwise observable as fragments in isolated datasets.³ This research fills these data gaps by observing known human graves with a variety of sensors over several years.

A 36-month experiment is underway which strives to address the following research questions: (1) Do human graves have one or more unique signatures that can be used for grave detection?; (2) Do differently sized graves affect detection capability?; (3) Does time affect detection capability?; and, (4) What combination of sensors/signatures is optimal for grave detection?

In February 2013, ten human bodies were buried in three differently sized graves in previously unused land at the ARF. The graves contain one, three, and six bodies, respectively. A fourth grave, the control, was dug and refilled without human remains and bears the same dimensions as the six-person grave. Baseline collections of terrestrial Light Detection and Ranging (LIDAR), terrestrial spectra, aerial LIDAR, satellite multispectral, and plant samples were made before burial. Since burial, incremental collections of terrestrial LIDAR, satellite multispectral imagery, and plant samples have been ongoing and will continue throughout the study. Post-burial data collection may be expanded to include aerial LIDAR, aerial hyperspectral imagery, and field spectroscopy in the near future. Ongoing data processing and analysis is being performed under the guidance of imaging scientists at GIST, a global leader in geospatial research.

Through this research, ARF and GIST researchers aim to meet the following objectives: (1) identify one or more detectable grave signatures through the fusion of data from multiple sensors over time; (2) evaluate the sensitivity of each sensor over differently sized graves; (3) evaluate signatures of graves created at different times; and, (4) assess the utility of the remote data fusion approach in both small- and large-scale event management processes.

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Remote Sensing, Mass Graves, Forensic Archaeology

D50 “Why is the Chief Here?!” The Importance of Pathologists Attending the Death Scene

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After attending this presentation, attendees will appreciate the benefits of pathologist participation at the death scene and understand the various roles of each professional in performing a death scene investigation.

This presentation will impact the forensic science community by communicating the benefits of direct scene attendance by the forensic pathologist both on the outcome of the investigation and on the management and service of the participants and customers of the death investigation.

Of the many duties the forensic pathologist performs, none is more important than attending death scenes. Most forensic pathologists delegate death scene inspection to others or attend only the rare high-profile scenes. Forensic pathologists who invest the time attending death scenes gain tremendous insight and professional benefits.

Forensic Autopsy: It is often stated that “*The forensic autopsy begins at the scene,*” yet attendance at death scenes by pathologists appears to be in decline. Whatever rationale is used for not attending death scenes is outweighed by the benefits. Viewing the body at the scene allows the forensic pathologist to obtain first-hand correlations of bodily injuries, body position, and evidence. Many times, law enforcement attending the autopsy do not have first-hand information of the specifics of the scene and even trained death investigators lack the depth of knowledge necessary to draw medical correlations between what is visible and what lies hidden. In addition, this critical step allows the forensic pathologist to save time performing the autopsy procedure itself and diminishes the possibility of misinterpretation of findings and rework.

Visualize the Death Scene: Observing the scene allows the forensic pathologist a unique perspective of spatial relationships not readily obtained with two-dimensional photography. The forensic pathologist can gain valuable insight into environmental conditions, locations, distances, injuries, and activities of the decedent.

Establish a Time of Death: Examining the decedent at the scene allows for a more accurate estimation of the time of death. The forensic pathologist can be much more helpful if the body is viewed prior to refrigeration.

Initiate the Chain of Custody: First-hand observations of evidence at the scene in relation to the body give the forensic pathologist substantial insight and advantage during autopsy. Even

the identification of evidence oftentimes requires expertise not typically held by law enforcement or trained scene investigators. Most training investigations can follow chain-of-custody procedures; however, the credibility associated with witnessing the actual collection gives any resulting court testimony additional reliability.

Debrief Law Enforcement: The scene debriefing with law enforcement by the forensic pathologist establishes the known factual information and assists (on scene) law enforcement in understanding medical opinions (i.e., the type and probable origination of wounds). This frequently defuses unnecessary guessing and avoids misconceptions on the part of law enforcement, before misconception becomes “a working hypothesis.” In addition, the pathologist can provide preliminary comments (which can be adjusted at autopsy) and give realistic time frames for autopsy performance and the scheduling of any additional studies that may be required.

Establish Credibility With Customers: Regardless of what may appear to be illogical, forensic pathologists have “customers.” Family members, district attorneys, law enforcement officers, jurisdictional administrators, political and religious leaders, the media, and others may enter the world of the forensic pathologist. Whether working for a Medical Examiner’s/Coroner’s (ME/C) office or serving as the ME/C, interaction with your “customers” in the unnatural environment of the death scene creates a lasting bond among professionals.

Most importantly, the family is especially approachable at the time of death. Personal contact with the family such as the exchange of a business card or condolence allows for the creation of a professional relationship with the family, their friends, and support systems. This pays dividends in difficult or delayed cases.

Review Operational Integrity: For forensic pathologists who also serve as chief medical examiners or coroners, the death scene is where many of the important components of the ME/C office come together. It is here that the forensic pathologist can witness professional interaction between death investigators, law enforcement, crime laboratory personnel, body transports, reporters, and others. The forensic pathologist can view the interaction and gain insight into the essential functions of the office and where improvements can be made.

Establish Credibility in the Courtroom: Oftentimes, the mere acknowledgment of having personally attended the death scene and participated in the scene investigation elevates the credibility of the forensic pathologist as an expert witness. The forensic pathologist is able to make statements based on first-hand observations, rather than secondary transfer of reported information. In many cases, the jury understands and appreciates the benefits of testimony provided by individuals who actually attended the death scene.

Death Scene, Forensic Pathologist, Investigation

D51 Multiple Sharp Force Injury Suicides: Common Features and Suggestions for Differentiation From Homicide Cases

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After attending this presentation, attendees will better understand the typical and atypical features of multiple sharp force suicides in Pennsylvania, as well as be able to apply criteria for differentiation between homicides and suicides.

This presentation will impact the forensic science community by providing a general description of multiple sharp

force suicides and practical suggestions for determining the manner of death at these rare scenes, with particular attention paid to the perspectives of the homicide investigator, the crime scene investigator, and the crime assessment specialist.

Sharp force suicides are rare, with cases that involve multiple sharp force injuries and/or multiple sharp force weapons composing an even smaller subset of these suicides. Due to the bizarre and sometimes gruesome nature of both the injuries and the immediate crime scene, it is understandable for investigative personnel to suspect the involvement of at least one other individual and to believe they are dealing with a homicide rather than a suicide. Careful attention to the scene, the body, and the gathering of information through interviews and other investigative queries is essential to distinguishing a homicide from a suicide in these cases.

Using four cases from Pennsylvania, this presentation will address typical and atypical features of sharp force and multiple sharp force suicides and the outdated criteria for differentiating the manner of death that have been contradicted in the literature. Central to this presentation is identifying and analyzing the features of these cases that determine how investigative personnel view and approach these scenes, as well as where suggestions for best practices can be made. Additionally, those attending this presentation will be able to see case analyses from the “investigative perspective” regarding forensic indicators of self-inflicted injuries, including blood pattern analysis, location of weapon and injuries, as well as the important background information necessary in these investigations, including medical history, personal history, and family dynamics. This presentation utilizes three multiple sharp force suicide cases that each exhibit both typical and atypical features, as well as a multiple sharp force homicide case that, upon cursory assessment, appears to have similar features to the three suicide cases. This presentation utilizes a multiple case study format to provide a holistic understanding of multiple sharp force suicides in an effort to aid others in the forensic community in making a correct determination of the manner of death in these case types.

Suicide, Multiple Sharp Force Injuries, Crime Assessment

D52 Stepping Into Squalor: Scene Investigation of Decedents Who Lived in an Extremely Cluttered or Squalid Residence at the Time of Their Death

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After attending this presentation, attendees will have a better understanding of the demographic and social history of decedents in Harris County who were living in an extremely cluttered or squalid residence at the time of their death.

This presentation will impact the forensic science community by providing new insight into the investigation of extremely cluttered death scenes and addressing common misconceptions of decedents who were living in that type of environment. The data presented may benefit first responders, investigators, pathologists, social workers, and other professionals by helping them to see beyond the squalid conditions and instead focus on specific clues inside the residence, thus being able to perform a proper and more thorough investigation.

Using definitions developed by Randy Frost, PhD and Gail Steketee, PhD, in their research on hoarding, 81 sets of scene photographs were examined for this study. All deaths occurred and were investigated in Harris County from 2009 to the present.

Scenes were determined to be extremely cluttered or squalid using the Frost/Steketee definitions. Information about the decedents was gathered from both the forensic investigators and pathologists reports. Cases selected were deaths of adults only (age 18 and older) and the death must have occurred inside of or on the property of an extremely cluttered residence. For this study, a residence was defined as a person's permanent living quarters and those quarters must contain a kitchen, bathroom, and living room or bedroom. Therefore, homeless individuals and those living in encampments were excluded. Children were also excluded because it is believed they would not have adequate control over their living environment.

Using the stated case criteria, decedent demographic data including age, race, and gender were compiled, as well as social history, including substance abuse, market value of the residence, whether family members were easily located, and the cause and manner of death (as determined by the pathologist). Scene photos were examined for patterns and trends and were also noticed in another investigation of extremely cluttered scenes.

Of the 81 deaths reviewed for this study, the majority of the decedents were White males and the average age of the decedents was 62 years. Those aged 62 years and older accounted for 50% of the 81 subjects. This age group was over-represented in this study compared to the population of Harris County whose residents age 62 years and over account for only 10.5% of the population. Most of the decedents were in an early-to-moderate stage of decomposition (65%) but, surprisingly, the majority of decedents had family members that either lived nearby or were in regular contact with the decedent (73%). Most of the decedents did not have pets (73%) and the majority of the decedents did not smoke cigarettes, use illicit drugs, abuse prescription medications, or use alcohol. However, of those who did use alcohol, most of the use was described as "heavy."

When investigating deaths in an extremely cluttered or squalid residence, it is beneficial for investigators, Emergency Medical Services (EMS), and police officers to be able to locate needed information quickly and safely. Though the scene appears overwhelming at first, some trends were noted which may assist investigators. For example, current medications, cell phones, purses, and other frequently used items were often found on top of piles which were closest to the bed, the door used to enter the residence, the television, and/or sitting area. There was no need for the investigator to risk his or her safety by sorting through the piles and debris.

This presentation will address common misconceptions regarding people who hoard and offer insight into successful navigation of extremely cluttered death scenes. This information will be of benefit to those who first respond to 911 calls, those investigating the death, and, ultimately, to the family of the deceased.

Hoarding, Forensic Investigation, Residence

D53 Pozzetto: Inside Behavioral Mazes of a Killer 27 Years Later

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After attending this presentation, attendees will have learned about one of the most violent episodes in the history of Colombia, not related to politics or guerilla and drug trafficking, featuring a unique murder.

This presentation will impact the forensic science community by showing, more than 20 years later, the meaning of a criminal action in light of criminal behavior analysis and behavioral evidence.

In Colombia, there have been many documented massacres. Many innocent people have suffered directly or indirectly from the actions of guerrillas, paramilitaries, drug traffickers, and politics. In these contexts, violence has become extremely brutal. But even after so many years of violence, one case remains in the memory of many as a very atypical case in Colombia.

In the first week of December 1986, one man, Campo Elias Delgado Morales, conducted one of the worst crimes that has been registered in Colombia. In a period of 24 hours in three different places, armed with a knife and a gun, he killed 28 people. He stabbed and burned his mother, shot her neighbors, bound and stabbed his English student and her mother, and, finally, after dinner, he shot at several customers of a restaurant named Pozzetto. After an exchange of gunfire with police, the murderer died. This case is called the Massacre at Pozzetto.

After ten months, the case was closed by order of a judge, who wrote in his decision: "...nor will this court stop to study the psychological causes that could induce Campo Elias Delgado Morales to this insane act...nor the reasons which led him to conceive of this macabre plan...entombed are the reasons for his actions." Words like "psychopath," "Vietnam veteran," "multiple personality," "PTSD," and even "serial killer" filled the pages of newspapers and news programs. The authorities were never able to establish a reason for what caused Campo Elías to commit murder before the case closed.

Today, more than 25 years after the event, the review of photographs, plans, autopsy reports, and case statements, in combination with the application of forensic science, may help explain some of the circumstances of the event. Ballistics evidence analysis and bloodstain pattern analysis "explain" some of the circumstances surrounding the death of a murderer, while the crime scene analysis and behavioral analysis provide a glimpse of the strong personal feelings of a murderer and the sexual motivation in some of the crimes. Finally, this study tries to explain what type of killer Campo Elias Delgado Morales was and the circumstances surrounding his actions.

Behavioral Evidence, Criminal Behavior, Murder

D54 The Evolution of Forensic Nursing Science Education and Mentorship

Joyce P. Williams, DNP, 10809 Stansfield Road, Randallstown, MD 21133*

After attending this presentation, attendees will be able to: (1) identify growth of forensic nursing science education, including the practice of forensic nursing; (2) analyze how well colleges and universities are performing in preparing forensic nursing scientists for advanced practice roles; (3) review the theoretical foundation for Advanced Practice Forensic Nursing; (4) demonstrate how to meet mentorship needs of forensic nurse practitioners; and, (5) communicate opportunities for, and barriers to, providing forensic nursing science education to international and U.S. students.

This presentation will impact the forensic science community by showing how quality advancement and improved systematic outcomes are anticipated outputs when building carefully articulated academic structures and organizational systems for the advancement of forensic nursing practice and leadership. Instrumental in the development is the inclusion of informational systems and technology to advance and transform health care. Maintaining a scholarly environment that serves

to educate advanced practice nurses in the forensic specialty is attainable with focused programs and dynamic models that include interprofessional collaboration as a critical component of the process, aligning with the essentials for doctoral nursing, and leading to improved patient and population health outcomes.

Statement of Proposal: The purpose of the presentation is to demonstrate the scientific underpinnings for practice essential to sustain the strength of forensic nursing science and to continue the path forward. The goal is to advance clinical scholarship and analytical methods for Evidence-Based Practice (EBP) among advanced forensic nursing practitioners.

Brief Synopsis: Academic learning environments for nursing scholars strive to create opportunities for improvements in science education while preparing the clinician for transition from theoretical knowledge and evidence-based research to clinical practice settings. Advanced Practice Nurses (APNs) with specialized forensic training are instrumental for setting the standard for EBP and optimal outcomes when evaluating victims of violence.

Summary: Doctoral training for the forensic APNs involves an interprofessional approach and is specifically designed to address injury in the person and populations.

Curricula schema includes methodology that meets the scope and standards for the nurse practitioner role and is strategically positioned for those working with individuals. The APN provides essential holistic care using a biopsychosocial approach to examine injuries suffered. Referrals and outreach is customary with the forensic interprofessional scientific community collaboration to attain best practice.

Content follows a pedagogic model comprised of three essential areas: (1) mapping course learning outcomes linking theory content, learning activities, and evaluation activities to the course learning outcomes; (2) identification of program outcomes with course learning outcomes (clinical judgment/scholarship in nursing practice, critical analysis of complex situations and practice systems, evaluation and application using conceptual models, theories and research to improve diversity in population health, systematic investigation of a focused clinical aspect to advance healthcare, analysis of the social, economic, political, and policy components contained within systems, manage leadership roles developing clinical practice models, health policy and standards of care, and integration of professional values and ethical decision-making in advanced nursing practice); and, (3) link Doctor of Nursing (DNP) expected competencies to course outcomes and content.

Forensic Nursing, Education, Forensic Science

D55 “Historical Hangover:” Backlogs as Hysteresis

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After attending this presentation, attendees will have knowledge of how cumulative inefficiencies in a system (such as a forensic laboratory) can lead to backlogs (unworked cases older than 30 days) and the conceptualizing backlogs as a type of hysteresis.

This presentation will impact the forensic science community by providing a basis for understanding the true source of backlogs, policy implications, and a new way of solving this persistent problem for forensic laboratories.

Hysteresis is the dependence on a system not only on its current environment but also on its past environment. If a given input alternately increases and decreases, the output tends to form

a loop. Loops may also occur because of a dynamic lag between input and output.

Rate-dependent hysteresis is where an input variable X cycles from X_0 to X_1 and back, the output $Y(X)$ may be Y_0 initially and a different value Y_2 on the return. The values of $Y(X)$ depend on the values that X passes through, but not on the rate of change of X . That is, backlog is backlog, regardless of how slowly (inefficiency) or quickly (disaster) it builds.

History matters in that predictable amplifications of small differences are a disproportionate cause of later circumstances; in the long run, this historical hang-over is inefficient. Inferior standards can persist simply because of the legacy they have built up. A more technical definition is when a path-dependent stochastic process has an asymptotic distribution that emerges as a consequential function of the process' history (a.k.a., a non-ergodic stochastic process); a fuller explanation follows.

A system will ratchet toward a state that is, to one degree or another, dependent, reversible, and inefficient. Because the system is asymptotic (history-dependent), it moves toward one or more attractors governed largely by externalities. Unless the system can control the allocation of internal resources, it will hit a less-than-efficient lock-state which will be costly to recover from. Exit and opportunity costs become barriers.

Forensically, this asymptotic state results in a backlog. The backlog is more than a simple inefficiency, but rather is the cumulative historical result of uncontrolled resource allocations and process change based on misperceptions and misinformation. If a productivity indicator is defined as Submissions — Reports ($S - R$), this creates a form of rate-dependent hysteresis demonstrating the rate of work that is being done under the current environment and backlog (unworked submissions over 30 days old) as a rate-independent hysteresis (because it has memory — the 30 days) indicating process efficiency. Some relation of the two should be an indicator of capacity — what could be achieved if the inefficiencies of the backlog were removed.

In the old phrase, “If you give a person a fish, they can eat for a day; if you teach them to fish, they can eat for a lifetime,” where the reality is, if $S - R$ does not equal backlog as measured by cases unworked after 30 days, then you may never reduce your backlog by working “harder” or “smarter.” The laboratory will have achieved a hysteresis state that may not be reversible due not only to internal processes but also externalities. The laboratory needs to be able to learn how to fish — just being given fish doesn't solve the systemic problem.

Viewing backlog as hysteresis provides a change in perspective, allowing for a systems approach to policy, methods, process improvement, and, eventually, backlog reduction.

Laboratories, Backlogs, Hysteresis

D56 Forensic Science Training Task Force: Recommendations for Training and Continuing Education

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The goals of this presentation are to provide the forensic science community with information on more effective and efficient ways to train new hires and to help identify the gap between education, training, and continuing education.

This presentation will impact the forensic science community by providing attendees information regarding the creation of recommendations on ways to effectively and efficiently

train new hires in forensic science agencies.

Training and continuing education was identified as a significant area of need within the forensic science community in several studies including the National Institute of Justice's (NIJ) *Forensic Sciences: Review of Status and Needs* (1999), the Technical Working Group on Education's (TWGED's) *Education and Training in Forensic Science: A Guide for Forensic Science Laboratories, Educational Institutions, and Students* (2004), and the *180-day Study Report: Status and Needs of United States Crime Laboratories* (2004). The 2009 release of the National Academy of Sciences (NAS) Report, *Strengthening Forensic Science in the United States: A Path Forward*, also addressed the issue of training and continuing education for all working forensic scientists. The NAS Report states that continuing education is "critical for all personnel working in crime laboratories as well as those in other forensic science disciplines..." The Report also states that "training should move beyond apprentice-like transmittal of practices to education based on scientifically valid principles...but there are no uniform recommendations on the content of training in the forensic science disciplines."

The Training Task Force, an initiative sponsored by the NIJ's cooperative agreement with West Virginia University, was created to develop and provide recommendations on what is needed to train new hires and to help identify the gap between education, training, and continuing education in forensic science. The creation of the Task Force was in line with the identified needs in the forensic science community that are being observed across the United States. The goal of forensic science agencies is to secure scientists who are skilled forensic professionals. It is important for scientists to be trained to competency to perform their assigned duties in a consistent manner from agency to agency, while keeping costs to a minimum. The task force included a variety of forensic professionals from different laboratories from both local and state agencies throughout the country. Professionals include laboratory directors, training coordinators, educators, and supervisors who are familiar with the importance of training new hires while keeping in mind the importance of cost effectiveness. The Task Force determined that training needs to be consistent between agencies so retention becomes less of an issue for those that have excellent training programs. The Task Force also identified gaps between formal education, training, and continuing education. Identifying these gaps will better the forensic community as a whole as training and continuing education programs can be more focused on the scientists' needs.

The goal of the Training Task Force was to draft recommendations and suggestions to the forensic community regarding training of new employees. Suggestions will include topics on how to make laboratory training programs more efficient by building on the educational background and current knowledge of the entry-level scientist who graduated from an accredited forensic science program. Since there are now standards of forensic science education at these accredited institutions, agencies can further support the programs while using the education as a benchmark. Building off the current knowledge of the new hire eliminates retraining in topics where competency has previously been demonstrated. For example, if two new employees are hired into the agency, one with several years of experience and the other right out of school, building off the current knowledge of the new employee will eliminate the time it takes to train the new hire with experience to competency.

Continuing Education, Training, NAS Report

D57 Drug Abuse and Misuse Contributing to Death in North Central Indiana

Matthew C. Wietbrock, BS, 629 N 6th Street, Lafayette, IN 47909*

After attending this presentation, attendees will learn of the contributions prescription drugs, illegal drugs, and alcohol have played in the deaths of residents of Tippecanoe County, Indiana, over the previous 20 years.

This presentation will impact the forensic science community by focusing upon the current state of prescription drug abuse and by displaying the frequency such drugs are encountered in deaths investigated by the Tippecanoe County Coroner's Office.

The prevalence of mind-altering substances compared to all cases investigated by the Tippecanoe County Coroner's Office will also be discussed. This data will also be compared to national trends.

Tippecanoe County is located 68 miles northwest of Indianapolis, and 124 miles southeast of Chicago, and is home to a population of 174,724. Tippecanoe County has a large manufacturing base including Caterpillar® Inc., Subaru®, Fairfield Manufacturing®, and Wabash National®, which are all represented in the county. Tippecanoe County is also home to Purdue University, a major research institution and the county's largest employer.

Like alcohol, drugs may lower inhibitions and, in some cases, induce high-risk behavior. This has long been recognized as prevalent in drug-involved cases, where the manner of death is found to be accidental, suicide, or even homicide. Minimally, the presence of drugs, including alcohol, are investigated for regularly. Alcohol, being readily available and easily abused, is often encountered in death investigations. Similarly, hard street drugs, such as cocaine, heroin, or synthetic drugs have also long been known to play significant roles in death investigations. Use and availability of these drugs, especially heroin, have continued to rise in the North Central Indiana Region.

Nationally, the number of prescription medication overdose deaths has continued to rise, and Indiana is no exception. Several high-profile celebrity deaths on the national stage have added to the public awareness. Tippecanoe County has experienced a rise in the use of opioids such as methadone and oxycodone for pain management. The potential for chronic abuse of these prescriptions has raised the concern of public health officials as has the potential for Opioids to be combined with benzodiazepines and/or alcohol, which often leads to harmful results. The Centers for Disease Control reported that overdose deaths related to the misuse of methadone have increased to account for one in three pain killer overdose deaths since 2009.

An examination of 20 years of death statistics from Tippecanoe County, Indiana, will yield the frequency in which illegal prescription drugs and/or alcohol have been detected in decedents' toxicological examinations, postmortem. This data will also display the frequency in which drugs and/or alcohol were detected as related to final determination of manner of death since 1993. This presentation will discuss the previous two decades and compare 1993-2003 to the previous decade. Trends will be displayed to educate attendees of current abuse patterns in north-central Indiana.

Death, Overdose, Prescription Drugs

D58 Comparative Analysis of the Public Health Role of Two Death Investigation Systems in Texas: Application of Essential Services

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After attending this presentation, attendees will understand the application of the ten essential public health services fulfilled by death investigation systems. Attendees will be able to appraise how two death investigation systems within the state of Texas Medical Examiner (ME) system and Justices of the Peace (JP) system fulfill these services. The results flow naturally to recommendations for standardization of practices and promotion of collaborative efforts.

This presentation will impact the forensic science community by serving as a key example of state assessment of medicolegal death investigation public health essential services.

The purpose of this research was to assess the policies and practices of medicolegal death investigation agencies in the State of Texas and to investigate differences between agency's perceptions of their role within their public health responsibilities. The public health responsibilities, which are applicable to all public death investigation systems, are summarized by the ten essential services.^{1,2} A survey was conducted in which JPs and MEs were questioned on their agency's policies and practices in regard to essential services provided. A recently developed and published 50-item instrument subdivided into ten essential service areas was used.¹ The essential services are summarized in these categories: monitor; diagnose and investigate; inform and educate; mobilize; policies and procedures; enforce; link; assure adequate standards; evaluate; and research. The study was a quantitative descriptive cross-sectional design in which nonparametric analysis was used to ascertain differences between groups.

Texas' 254 counties are divided into 11 public health regions, which are served by 12 MEs and approximately 723 JPs. The 12 medical examiners' offices and approximately 723 JPs were the participants of interest. The respondent sample size for analysis was composed of 10 (83%) ME offices and 112 (15.5%) JPs. The study showed significant differences in the execution of the majority of roles and functions, both across respondents and between the two groups of medicolegal death investigation agencies in providing essential services. Specifically, this significance was pronounced when examining responses to items addressing patient safety and healthcare treatment-related deaths and how hospital administrators and forensic providers identify, investigate, and classify these deaths.

This presentation will inform attendees about the findings of this study and provide recommendations for the standardization of medicolegal death investigation in the state of Texas.

References:

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2. National Public Health Performance Standards Program [NPHSP]. *Local public health System performance assessment instrument*. Retrieved on July 31, 2013 from <http://www.cdc.gov/od/ocphp/nphpsp/TheInstruments.htm>

Forensic Sciences, Death Investigation Systems, Essential Service

D59 Comparing Electronic News Media Reports of Potential Bioterrorism-Related Incidents Involving Unknown White Powder to Reports Received by the United States Centers for Disease Control and Prevention and the Federal Bureau of Investigation Reports: USA — 2009-2011

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After attending this presentation, attendees will understand that "white powder" incidents occurred in practically every state and region in the U.S., that these unknown "white powder" incidents also occurred in various public or private places, and that one cannot assume that these unknown white powders are harmless or that they pose no public health threat or threat to national security.

This presentation will impact the forensic science community by showing the relevance to various public health and private emergency responders, criminal investigators, and forensic laboratories. The presentation highlights that potential bioterrorism-related incidents related to white powder continue to occur, and that these incidents require an integrated response with personnel from various disciplines.

Using various search engines, the researchers performed internet searches for "unknown white powder" that were reported from June 1, 2009, and May 31, 2011. The white powder incidents that were reported online by news media were reviewed and compared to the white powder incidents reported to the Centers for Disease Control and Prevention (CDC) and the Federal Bureau of Investigation (FBI) during the same time period. The following types of information were abstracted: report date; state of incident; specific location of incident; identification of the unknown white powders; emergency responders involved; and FBI involvement. Using Excel® 2003, an electronic database was constructed, and a descriptive statistical analysis was performed using SPSS® 17.0.

In this study, there were 297 white powder news media reports from 43 states and the District of Columbia. The CDC received 538 white powder reports, and the FBI Weapons of Mass Destruction (WMD) reported 384 incidents during the same period of time. Out of the 297 white powder incidents reported by the electronic news media, only five were also reported to CDC. Out of the 538 white powder incidents reported to CDC, only five were also reported by the electronic news media. In other words, there were only five white powder incidents that were reported both to CDC and reported by the electronic news media during this two-year period. For the FBI data, this study found that "region" is the smallest geographic parameter that is available for analysis. It was not possible to correlate white powder incidents from the three sources at the state level. Therefore, this study was not able to determine how many incidents were reported to the FBI and similarly reported to CDC and/or covered by the electronic news media.

Many of the online media-reported domestic incidents came from Florida (n=31, 10.4%), California (n=26, 8.8%), Texas (n=23, 7.7%), New Jersey (n=21, 7.1%), New York (=19, 6.4%), Alabama (n=13, 4.4%), Utah (n=13, 4.4%), Virginia (n=12, 4.0%), Washington, DC (n=10, 3.4%), and Massachusetts (n=10, 3.4%). Many of the reported electronic news media incidents occurred in law enforcement offices, courthouses, and jail/correctional

facilities (n=38, 12.7%). Government locations other than those three mentioned previously (n=80, 26.9%) included federal building mailrooms, state offices, city hall, Internal Revenue Service, U.S. Attorney's Office, Senator's office, Division of Motor Vehicles, Veterans Affairs Administration, U.S. Post Office, military call center, State Attorney General's office, Social Security Administration, and others.

The majority of news reports (n=223, 75.1%) did not mention the final identity of the suspicious white powders. Those that were identified included sugar/artificial sweetener (n=10, 13.5%), flour (n=9, 12.2%), baby powder (n=5, 6.8%), and talcum powder (n=5, 6.8%). None of the substances were identified as a bioterrorism agent.

Joint public health and FBI investigations were mentioned in some of the electronic news media reports (n=66, 22.2%). Other responders that were commonly mentioned in the electronic news media reports were fire departments (n=56, 18.9%) and police departments (n=51, 17.2%) which could be at the state or local levels.

This analysis demonstrates that none of the three entities collected data for all white powder incidents. Furthermore, results of this study serve as a reminder that white powder incidents require an integrated response with personnel from various disciplines: public health; law enforcement; and Fire/emergency Medical Services (EMS). Collaborative efforts between law enforcement and public health continue to be enhanced, including routine sharing of information. This data sharing is critical to assure that any city or state is prepared to respond to perceived or actual biological threat agents in the future.

Bioterrorism, Unknown White Powder, Anthrax

D60 Status of the Scientific Working Group on Bloodstain Pattern Analysis (SWGSTAIN)

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After attending this presentation, attendees will have an understanding of the objectives and purposes of the Scientific Working Group on Bloodstain Pattern Analysis, as well as the potential impact the guidance documents produced by this working group have on the discipline of bloodstain pattern analysis.

This presentation will impact the forensic science community by reviewing the existing SWGSTAIN guidance documents and online resources, as well as updating them on guidance documents currently being drafted by SWGSTAIN.

In 2002 the Federal Bureau of Investigation (FBI) Laboratory coordinated a meeting to explore the idea of a scientific working group related to Bloodstain Pattern Analysis (BPA). An international core group of 16 recognized BPA experts affirmed the need for a scientific working group in BPA — SWGSTAIN. Currently, the membership of SWGSTAIN includes 29 recognized bloodstain pattern analysts who represent law enforcement agencies, laboratories, and private practitioners in North America, Europe, New Zealand, and Australia. In 2009, the Midwest Forensics Resource Center assumed the leadership role of SWGSTAIN from the FBI Laboratory.

The mission of SWGSTAIN is to promote and enhance the development of quality bloodstain pattern analysis practices through the collaborative efforts of governmental forensic laboratories, law enforcement, private industry, and academia. Currently, the SWGSTAIN membership is split into six standing subcommittees: (1) Taxonomy and Terminology; (2) Training and

Education; (3) Quality Assurance; (4) Legal; (5) Research; and, (6) Business and Strategic Planning. Each of these subcommittees is working on specific tasks deemed necessary by the entire working group. During the biannual meetings, each subcommittee presents status updates to the entire working group. Once a committee is nearing the completion of a guidance document, the document is then distributed among the entire group for comment. This often occurs multiple times prior to the documents being published for public comment. Documents are not released for public comment until the draft is passed through the membership and then through the SWGSTAIN Executive Board. After the deadline for public comment has passed, the issuing subcommittee considers each comment and acts on it. The document then cycles through the voting process.

The role of each subcommittee will be discussed along with its respective published guidance documents and existing online resources, which include an extensive BPA bibliography and an admissibility resource kit. The status of in-progress projects will be presented. SWGSTAIN has targeted/recommended a dozen BPA research priorities

In addition, SWGSTAIN is working to be proactive toward current issues impacting their discipline. The work of SWGSTAIN is on-going and relies upon the input of BPA practitioners. The current documents that are available for public comment as well as the completed guidance documents can all be found on www.swgstain.org.

Bloodstain, Bloodstain Pattern Analysis, SWGSTAIN

D61 Adult Suicides in Northern Virginia: A Look at Trends From July 2012 to June 2013

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After attending this presentation, attendees will learn about the suicidal trends among adults (ages 22 and older) in the Northern Virginia (NoVA) area which include: methods of suicide; the correlation of suicide with substance abuse; mental illness; cohabitation with family; and the impact of physical ailments or diseases on suicide. Attendees will also learn that these factors can be specific to geographic areas and local populations. Knowledge of suicidal trends in one's own jurisdiction is pertinent for a thorough and accurate investigation. Having the ability and foresight to investigate a case with this information in mind will assist in capturing comprehensive and precise data for future research in the area of suicide.

This presentation will impact the forensic science community by raising awareness of suicidal trends among adults in the Northern Virginia region. By comparing data of this study to state and national populations, the importance of regional and national tendencies is revealed. This data can be used as a springboard for future research on suicide trends in even more specific populations such as young adults, the elderly, returning war veterans, and those recently released from jails and institutions, with the goal of encouraging death investigators to refrain from generalizing suicide cases and to investigate each case individually.

This study proposes that methods of suicide and the correlations between suicide rates, the use of drugs and alcohol, psychiatric diagnoses, medical ailments, and living arrangements with family vary according to region. The information gathered provides a tool to all death investigators in raising awareness that the region of death may affect the trends associated with suicide.

The information for this study was obtained with the aid

of statistics from the Virginia Violent Death Reporting System (VVDRS). Decedent age, sex, race, jurisdiction, and cause of death were gleaned from information reported through the VVDRS via the Northern District Office of the Chief Medical Examiner. Other pertinent information such as history of drug or alcohol abuse, living arrangements, history of mental illness, marital status, and sub-fatal agency was gathered through individual case investigation. An analysis was completed on 251 adult suicide cases which occurred in the Northern Virginia jurisdiction from July 1, 2012, to June 30, 2013. Correlations were created using a Microsoft® Excel® spreadsheet which analyzed the average age of adult decedents, common methods of suicide, jurisdictions, trends regarding sex and methods, the impact of mental illness, medical diseases, and substance abuse, and trends regarding adults who become dependent and move back into their parents' homes.

Adult, Northern Virginia, Suicide

D62 Psycholinguistic Credibility Assessment of News Reports About a 142-Year-Old Mass Murder

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After attending this presentation, attendees will understand how the principles of psycholinguistic credibility assessment may be applied to news or literary accounts of major events to evaluate the veracity of the writers.

This presentation will impact the forensic science community by demonstrating how credibility assessment may be used on historical news and literary documents to determine the veracity of victims and witnesses even when formal statements were never made or were lost to history.

On the morning of November 5, 1871, a Concord stage coach en route from Wickenburg, Arizona Territory (AT), to Ehrenburg, AT, was ambushed about eight miles west of Wickenburg. The driver and five passengers were killed during the ambush, but Mollie Sheppard, the only female passenger, and William Kruger, a civilian Army clerk, though severely wounded, managed to escape. Sheppard later reportedly died from her wounds.

Suspicion immediately fell on Yavapai Indian warriors from the nearby Camp Date Creek Reservation, based on physical evidence from the scene and the statements of Kruger and Sheppard. However, the nature of the attack differed considerably from the Indians' usual *modus operandi*, and Charles Genung, a prominent and well-respected local rancher, attempted to lay the blame on Mexican bandits. Moreover, Kruger's and Sheppard's accounts of the massacre were called into question; Sheppard's because she was a prostitute and therefore disreputable, and Kruger's because of his unflattering portrayal of the local Army Commander and some of the citizens of Wickenburg.

Though neither Kruger nor Sheppard made formal written statements, Kruger wrote a lengthy letter to a relative of one of the dead passengers, which was published in the Boston and New York papers. In it, he provided details of the ambush and complained of poor treatment from the local Army installation at Camp Date Creek, AT. In a later interview by a reporter in San Francisco, he provided additional details of the ambush and complained about the hasty and inadequate burial of one of his fellow travelers. His published accounts brought a flurry of angry responses from the officers of Camp Date Creek and the *Arizona Miner* newspaper branded him "a contemptible liar and slanderer."

Kruger's letter and San Francisco interview were analyzed using the principles of psycholinguistic credibility assessment previously reported as well as the rebuttal letters from three Army

Officers and the article in the *Arizona Miner*.¹ Kruger's letter was found to be structurally consistent with a truthful statement and conveyed genuine emotion when reporting his difficulty in securing treatment for himself and his companion. The letters of the Camp Date Creek Commander and Adjutant both contained many indicators of deception by reporting carefully unattributed facts, such as, "I am informed by an officer..." and "...So at least I am informed by reliable parties..." The third letter, from the post doctor, is generally consistent with a truthful report, but very clearly conveys a sense that it was written at the order of the Camp Commander, and that the only factor that delayed him in ordering Miss Sheppard off post was the doctor's concern over the swollen and infected nature of her wounds.

Kruger's published interview in San Francisco likewise is structurally consistent with a truthful statement and again conveys emotion regarding his horror upon observing the bones of one of his traveling companion sticking up from a shallow grave beside the road. The denial in the *Arizona Miner* was not suitable for psycholinguistic analysis, but, in this instance, additional facts from other contemporary sources tended to substantiate Kruger's report.

In conclusion, psycholinguistic credibility assessment may be applied retroactively to any first-hand or accurately transcribed report of a historical or contemporary event for the purpose of determining the truth and veracity of that report.

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Credibility Assessment, Historical Documents, Cold Cases

D63 Trends in Suicide: An 11-Year Retrospective Review From the Wayne County Medical Examiner's Office With Emphasis on 0-18 Age Group

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After attending this presentation, attendees can expect to learn about trends in child suicide observed from Wayne County Medical Examiner's Office during an 11-year period, where 5.1% of all suicides involved people aged 0-18 years.

This presentation will impact the forensic science community by influencing the direction of where future emphasis may be placed as it relates to child suicide prevention as five out of seven cases with prior suicide attempts had histories of depression. A need for increased therapy and follow-up may be warranted in some cases as evidenced by child suicide data reviewed by the Wayne County Medical Examiner's Office.

In the United States, suicide is the leading cause of injury mortality for all age groups combined surpassing motor vehicle crashes.¹ To assess trends in suicidal persons in the interest of prevention, a retrospective study was conducted at the Wayne County Medical Examiner's Office, with emphasis on 0-18-year-olds. A total of 2,083 suicide cases for all age groups were reported between the years 2000 and 2011. There were 107 (5.1%) childhood suicides involving people between ages of 0 and 18 years old. Ninety-eight child suicides occurred within teenage groups, constituting 4.7% of all suicides. More males (74 cases) compared

to females (33 cases) committed suicide in childhood. There was an equal incidence among White and Black youth suicides with 49 cases each. Other races comprised 9 cases. The leading method was hanging (65 cases), followed by gunshot (35 cases), drugs (4 cases), and multiple injuries (3 cases).

Outside the United States, a study from Jamaica found that hanging was the method of suicide in the majority (96%) of adolescent suicide deaths.² The U.S. Centers for Disease Control found in a 2013 study that most youth suicides were by hanging/strangulation/suffocation.³ Throughout the state of Michigan, the leading mechanism of suicide for all age groups is firearms. Firearms attribute to death in most suicides across all age groups combined, this study found. It could be postulated that prevalence of firearms may contribute to the occurrence rate of gunshot suicides versus hangings in youth. The adjusted suicide rate of 2.1 in this study is similar to the average adjusted suicide rate across the state of Michigan. The trend of increased teen suicides observed nationwide (8.2 for age group 15-19 years of age, as compared to 1.5 for age group 10-14 years of age) is supported by this study where the majority of the child suicide deaths were of teens.

Comparisons can be made to adult data, where the highest incidence of suicide was observed among adult Whites (1,560 cases), followed by adult Blacks (513 cases), and other races (49 cases). The 31- to 40-year-old age group conveys the most suicides (469 cases) in this study. A suicide note (written/verbal/text) was left behind by 20 of the child suicide cases (19%). Seven cases of suicide ages 18 years of age and under had a history of prior unsuccessful suicide attempts (6.5%). Twenty-nine cases had a medical history of depression, suicide ideation, developmental delays, or substance abuse (27%). There was reported history of bullying in one case and rape in another. Five people out of seven with previous suicide attempts had a history of depression, further signifying the need of interpersonal therapy and follow-up.

References:

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3. Karch DL, Logan J, McDaniel DD, Floyd CF, Vagi KJ. Precipitating circumstances of suicide among youth aged 0-17 years by sex: data from the national violent death reporting system, 16 states, 2005-2008. *J Adolesc Health*. 2013;53:1:suppl:51-3.

Child, Suicide, Trend

D64 The Scientific Working Group on Dog and Orthogonal Detector Guidelines (SWGDOG)

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After attending this presentation, attendees will understand how improved training materials and the establishment of best practices for detection teams is improving interdiction efforts and courtroom acceptance of dog alert evidence as well as the importance of creating an accreditation commission, the

International Commission on Detector Dogs (ICODD).

This presentation will impact the forensic science community by providing a better understanding of how the Scientific Working Group on Dogs and Orthogonal Detector Guidelines (SWGDOG) and accreditation through ICODD is improving the consistency and performance of deployed detector dog teams and their optimized combination with emerging electronic detectors.

SWGDOG's goals are to discuss and share ideas regarding methods, protocols, quality assurance, education, and research to bring together organizations and/or individuals actively pursuing relevant analysis methods to cooperate with other national and international organizations in developing relevant standards and to monitor and disseminate research and technology related to the discipline.

SWGDOG best practices have been developed by a membership of respected scientists, practitioners, and policy makers representing diverse backgrounds. SWGDOG has been cooperatively funded by the National Institute of Justice (NIJ), Federal Bureau of Investigation (FBI), Department of Human Services (DHS), and Technical Support Working Group (TSWG) since 2005. This project was undertaken as a response to concerns coming from a variety of sectors including law enforcement and homeland security regarding the need to improve the performance, reliability, and courtroom defensibility of detector dog teams and their optimized combination with electronic detection devices.

The approval of each subcommittee best-practice document takes a minimum of six months to complete including a two-month period of public comments. The ten SWGDOG subcommittees are as follows: (1) unification of terminology; (2) general guidelines for training, certification, maintenance, and documentation; (3) selection of serviceable dogs and replacement systems; (4) kenneling, keeping, and health care; (5) selection and training of handlers and instructors; (6) procedures on presenting evidence in court; (7) research and technology; (8) substance dogs (agriculture; arson; drugs; explosives; human remains; contraband; pest; currency; firearms); (9) scent dogs (non-specific human scent wilderness area search; location checks; article search; scent identification line-ups; live people in disaster environments; track trail people based on last known position; pre-scented canines aged trail; live people in avalanche); and, (10) outreach and education

The success of SWGDOG is dependent on the 55 SWGDOG members as well as the numerous external members within the working dog community who take the time to provide detailed commentary during the public comment stages. To date, there are 39 approved guidelines within 436 pages of resources. SWGDOG is a catalyst in prioritizing research and development in both canine and orthogonal detector areas in direct support of local law enforcement activities. The current success of SWGDOG is being made manifest by a shift of some national canine organizations to adopt the approved SWGDOG best-practice guidelines. Furthermore, the documents prepared thus far by SWGDOG members have been used to support the requirements of Homeland Security Presidential Directive -19 (HSPD-19), *Combating Terrorist Use of Explosives within the United States*.

Detector Dogs, Best Practices, SWGDOG

D65 Four-Legged Victim or Violent Predator: Can Necropsies Aid in Court Cases?

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The goal of this presentation is to provide insight into the legal ramifications and address the benefits of necropsies in criminal and civil cases. The forensic community, especially law

enforcement, attorneys, and veterinarians, can appreciate the need for necropsies, not just for legal means, but for educational and training purposes. Attendees will be able to enhance their investigative knowledge and take back some alternatives in handling animals at crime scenes. Attendees can assist with implementing new policies, procedures, and specific training needed for law enforcement and first responders in order to prevent or reduce the number of animal shootings.

This presentation will impact the forensic science community by providing definitions and will outline the number of animal deaths by law enforcement including audio and video footage of the shootings. The animal owners are painstakingly trying to figure out why their loved ones were, in their minds, unnecessarily shot. Police procedures and policies, as well as local and state laws, will be discussed. One or two unusual cases involving a necropsy of a goldfish and/or cockatoo and how this aided the court case will also be presented. Attendees will have the opportunity to look at various state laws already in existence regarding animals and their rights.

The world is changing and becoming more populated with different cultures, mores, and practices. Crime scenes are changing as well with people committing unimaginable acts of violence on other people and animals. Society has become private voyeurs with the increase in crime scene shows, YouTube® videos, and documentary-style programming like the ID channel's *Wives with Knives*, *Crime Scene Creatures*, *Animal Planet*®, and Oxygen® Channel's *Snapped*. The community is fascinated with law enforcement activities and appear to come running with video recorder in hand whenever they see police engaging in their daily duties.

The United States dog and cat population reached approximately 145 million in 2012, with about 56% of all United States households having a pet. Approximately 70 million households have dogs and 74.1 million households have cats. Birds, horses, specialty, and exotic pets are in about 17% of households. The types of scenes that the forensic community investigates are typically in residences, office buildings, on roadways, in vehicles, open fields, and inside containers. Most scenes involve only humans; however, increasingly family pets, specifically dogs and cats, are at the scene and need to be considered and managed during the investigation. Since 2010, approximately 228 dogs have been shot by law enforcement in the Houston area, resulting in the death of 142 dogs. Harris County is not the only area that has a problem with law enforcement shooting animals at scenes. There is a need for the forensic community to give animals the same respect and due diligence given to human remains, as these family pets may have grieving loved ones left behind.

Animals, Necropsy, Shooting

D66 The Whistler Sled Dog Case: The Exhumation of Clandestine Canid Mass Graves in British Columbia, Canada

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WITHDRAWN

D67 The Role of the Mexican Consulate in the Identification of Human Remains of Those Who Perish in the Desert Attempting to Cross the Mexican Border Into Arizona

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After attending this presentation, attendees will gain an understanding of the critical role played by the Mexican Consulate in conjunction with the Maricopa County Medical Examiner's Office with regard to the identification process of decedents believed to be Mexican nationals who have died attempting to cross the Mexican border.

This presentation will impact the forensic science community by demonstrating the increased success rate of a comprehensive approach between the Maricopa County Medical Examiner's Office and the Mexican Consulate during the identification process of human remains of those who succumb to the elements while attempting to cross the border from Mexico to Arizona.

The Mexico-Arizona border is crossed by thousands of immigrants every year. Due to many dangerous elements in the Arizona desert, many who attempt to cross the border do not make it alive. Arizona is ranked third in the United States with regard to unidentified human remains and Maricopa County is ranked second in the state of Arizona. Many of the unidentified decedents in Maricopa County, Arizona, are those who subsequently die while attempting to cross the border from Mexico to America due to the heat and menacing desert terrain. The effects of these environmental elements on the remains, combined with the fact that those crossing the border purposely try to keep their identification information hidden, make the identification process extremely challenging. There is often personal property found with the decedent that indicates they had been trying to cross the border from Mexico. Personal items that are found include Mexican currency, reading materials in Spanish, documents, and, occasionally, a Mexican identification card is also found.

The Unidentified Coordinator for the Medical Examiner's Office will review the personal property and provide a detailed summary for the Mexican Consulate representative that includes demographic information about the remains. The extreme heat and the effects of environmental elements typically leave human remains found in the desert unidentifiable, both visually and by identifying features, as most are primarily skeletal remains. Due to the condition of the remains, the identification can only be made through dental records/radiograph, medical implant radiographs, and DNA; fingerprints are not usually an option with skeletal remains.

The Mexican Consulate plays a major role in the identification process as they are able to search their own missing persons information and compare it to the demographic information obtained from examinations conducted by forensic odontologists and anthropologists at the Maricopa County Medical Examiner's Office. The Mexican Consulate representatives are able to compare information from family that might have called the Mexican Consulate worried because they have lost contact with their family member and had knowledge they were going to attempt to cross the border. Due to language barriers and cultural beliefs, the Mexican Consulate is able to obtain the information from the family in Mexico and relay to the Unidentified Coordinator at the Medical Examiner's Office in order to confirm the identification and attempt to adhere to cultural beliefs. The representatives for the Mexican Consulate also assist with sending DNA buccal swab kits to family in Mexico, verify Mexican ID cards are valid, obtain fingerprints

cards from Mexico, and obtain medical or dental imaging and other documents.

This study presents case scenarios and detailed information about the assistance the Mexican Consulate provides to the medical examiner's office when working unidentified human remains cases and finding family who live in Mexico. Coordinated multidisciplinary efforts are increasing the identification rate as a direct result from collaboration between the Mexican Consulate and the Medical Examiner's Office in Maricopa County, Arizona.

Identification Process, Mexican Consulate, Unidentified Remains

D68 Illegal Border Crossers in Arizona and California: A Comprehensive Study and Comparison of the Number of Deaths, the Causes, and the Related Factors and Patterns During the Last Decade

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The goal of this presentation is to communicate a study that was conducted of the continuance of illegally crossing the border and the measurement of associated deaths from 2007 through 2012 compared to the previous six years. Two main locations were used in this research: San Diego County, California, and Pima County, Arizona. Statistics were generated by studying the total number of deaths in each location by month and year, breaking the information down by gender and age, and discerning the six major causes of death. Comparisons in all areas were made between San Diego and Pima Counties based on this research, as well as collectively and generally compared to similar statistics from 2000 to 2006.

This presentation will impact the forensic science community by providing attendees with a better understanding of the details and the magnitude surrounding the deaths of illegal border crossers into the United States from Mexico. The numbers and the main causes of death will be investigated, as well as how these statistics compare to the information available from 2000 to 2006. Hopefully, this research will provide a platform for comparison and evaluation of the effectiveness of prior and current border-crossing initiatives, and will also provide a tool with which counties can better plan their budgets in regard to the costs of identifying those who die.

Ideally, this study would have included statistical information from all the other counties in these two states which border Mexico (Imperial County in California; Yuma, Santa Cruz, and Cochise Counties in Arizona), but due to variances in database systems, different sets of criteria used for recording information, and/or lack of information available, this study was unable to incorporate statistical data from those counties. During the time frame of this study, there were a total of 61 causes of death recorded on the data received from the Medical Examiner's offices of San Diego and Pima Counties for those who died while in the process of crossing the border. Many were the same basic cause of death; some were documented with more information or multiple causes were included. For purposes of this study, the six most common "causes of death" were focused on, including the three following categories: other, undetermined, and pending.

This research indicated that although border-crossing initiatives have been in place, modified and expanded upon over the years, the overall number of deaths have not decreased, but have become more numerous. Reasons for the increase of deaths in some locations can be directly correlated to the decreases in deaths in other locations. The causes of death have not changed significantly, but the increasing costs of autopsies have brought about changes in the way the postmortem examination costs are covered.

Border Initiatives, Border-Crossing Deaths, Medical Examiner

D69 An Unusual Homicidal Cervical Stab Wound With Complete Section of the Spinal Cord Examined by Postmortem Angio-Computed Tomography

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After attending this presentation, attendees will understand the interest of a Multiphase Postmortem Angio-Computed Tomography (MPMACT) examination in cases of numerous and multidirectional injuries.

This presentation will impact the forensic science community by providing an example of an unusual case of homicidal stab wound with complete transversal section of the cervical spinal cord.

Introduction: Cases of spinal cord injuries secondary to stab wounds are rare in the literature. In North America, spinal cord traumatism represents 2.6% of all the traumas and only 1% of them is secondary to stab wounds. In most of cases, those injuries are secondary to ballistic trauma. In a South African study consisting of 450 cases of stab wounds, the complete cervical spinal cord represented only 4.5% of the cases. Furthermore, lethal cases are rare and, in most cases, victims survive and present neurological sequels like tetraplegia or Brown-Sequard syndrome.

Case: The case of a 26-year-old woman killed by her boyfriend secondary to numerous stab wounds is presented. An MPMACT was performed before the medicolegal autopsy. The body was prepared with a surgical cannulation of the femoral vessels. After a non-enhanced Multi-Slice Computed Tomography (MSCT) exploration, a controlled perfusion device (Virtangio® machine) was used with paraffin oil mixed with a special contrast agent (Angiofil®), allowing for three time-different acquisitions (arterial, venous, and dynamic).

This exploration revealed multiple stab wounds (facial, thoracic, and cervical) with two lethal cervical lesions. One of the cervical lesions was located at the upper posterior part of the neck with a horizontal trajectory and complete transversal section of the cervical spinal cord between the first and the second cervical vertebrae. This lesion was accompanied with venous and arterial sections. The second cervical lesion was anterior, located on the

right side at the base of the neck and presented along its trajectory an injury of the right internal jugular vein and a section of the right transverse apophysis of the seventh cervical vertebrae. At the facial level, fractures of the right mandible were noted along the path of one stab wound. At the chest level, a right hemo-pneumothorax was noted with one scapula bone lesion secondary to one posterior stab wound.

Discussion: Only one similar case of complete transversal section of the cervical spinal cord with a lateral and horizontal trajectory between C1 and C2 was found. In this case, Rubin *et al.* enhanced the usefulness of radiologic investigation in instances of spinal injuries. Furthermore, exploration of the spinal cord is always a particular challenge at the autopsy. In this case, the dissection was easier because of the imaging conclusions and the realization of different-time acquisitions was essential for detection and comprehension of the origin of the bleeding. The precise localization of the injured vessels at the upper cervical posterior area was possible only with imaging. Moreover, MPMCTA permitted better *in situ* lesions' illustration and documentation.

Only the external examination permitted an exhaustive description of the different wounds. This unusual case due to stab wounds illustrates the complementarity of the MPMCTA and autopsy in instances involving those types of lesions.

Stab Wound, Homicidal, Postmortem Angio-CT

D70 Exploration of Isotope Ratio Mass Spectrometry as a Method for Thread Analysis

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The goal of this presentation is to present Isotope Ratio Mass Spectrometry (IRMS) as a prospective method in thread analysis, its use beyond the differentiation of white cotton threads, and the effect several factors have on the isotope ratios of threads.

This presentation will impact the forensic science community by demonstrating the potential of using IRMS for the analysis of various threads.

Clothing threads can be used as crime scene evidence if there are techniques that can compare the threads to their sources. Current methods use infrared spectroscopy and microscopy to analyze color, chemical composition, and a cross section of threads for comparison to the thread source. Different white cotton threads cannot be differentiated if they have similar color and composition, assuming that no biological evidence exists. IRMS is promising as it has been shown to be able to detect significant differences in carbon, oxygen, and hydrogen isotope ratios of white cotton threads from different sources. These differences in isotope ratios result from the cotton crop sources undergoing varying degrees of fractionation as a result of their unique environments; but it is not yet fully known what other factors affect isotope ratios of clothing threads.

The goal of this research is to investigate the potential of this method for other natural threads and determine the extent to which isotope ratios are impacted by factors such as dyeing, sweating, bleaching, and thread location throughout an article of clothing. Samples were collected from various t-shirts, silk fabrics, and jeans. Two used multicolored cotton shirts from different manufacturers were used in order to study commercial dyeing and a new, white cotton shirt was used to study household dye and the effects of bleaching it. Another new white cotton shirt was

also used to explore how the combined effects of sweating and bleaching influence the isotope ratios of cotton threads. Four silk fabrics with different stitching were acquired to expand this study to other natural threads. The shades of blue in three different pairs of jeans were also studied to investigate homogeneity of jean threads and the effect the amount of dye has on isotope ratios.

These clothing samples were acquired and combusted in either an Elemental Analyzer (EA) or high Temperature Combustion/Elemental Analyzer (TC/EA). A continuous flow isotope ratio mass spectrometer was used to analyze carbon isotopes in carbon dioxide gas, which is formed in the EA, and oxygen and hydrogen isotopes in carbon monoxide and hydrogen gas formed in the TC/EA. The differentiability of isotope ratios of two threads was determined using two-sample t-tests with a 95% confidence level. Visualization of the comparison between different threads using all three isotope ratios is achieved through three-dimensional trivariate plots.

Threads of different color in the multicolored shirts varied in their differentiability. It was determined that for one of the shirts, fewer than half could be differentiated using IRMS, while for the other, nearly all colors could be differentiated from each other. The multicolored shirts also showed differences in isotope ratios between different panels. For the dyed white cotton shirts, the threads showed no significant change in their isotope ratios from the undyed source. This implies that the differentiability among colors found in the multicolored shirt is likely due to cotton of different sources. When the dyed threads are bleached, the threads are differentiable from both the dyed and undyed threads, indicating that bleach changes the chemical composition of the threads instead of reversing them to their original, undyed states. Sweat can also significantly change the isotope ratios of cotton threads and make the altered threads differentiable from the original. Silk threads with different stitching are found to have significantly different isotope ratios and thus are differentiable from each other. White threads in one pair of jeans were found to be differentiable from the dyed threads using carbon isotope ratios. These findings enhance IRMS analysis for cotton threads by elucidating the effects of dyeing and bleaching on their isotope ratios and expand the scope of this method to other natural threads.

Isotope Ratio, Mass Spectrometry, Thread Analysis

D71 Mentor-Mentee: The Challenge to Create Our Future Scientists

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After attending this presentation, attendees will have a better understanding of the complex relationship between the mentor, the trainee, and the goals of the laboratory.

This presentation will impact the forensic science community by providing mentors with a better idea of the goals and aspirations of today's next generation of forensic scientists. Students will get a glimpse into the real world of the forensic lab and what awaits them after leaving the ivy-covered walls.

The world of criminalistics has seen a sea change brought about by external forces in the recent past. Shows like *CSI* and *The New Detectives* have made forensic science glamorous which has led to an increase in the number of students who want to study forensic science. This has led to an explosion in the number of colleges offering forensic science degrees, though the quality of

these programs varies widely. These students are ready to go to the crime scene, collect the evidence, analyze it, interview the suspect and witnesses, and make the arrest. At the same time, the field has had to deal with the accusations and recommendations of the National Academy of Sciences (NAS) Report. Meanwhile, labs that are accredited are dealing with the increased training requirements of ISO 17025. A great forensic lab does not merely train technicians, but educates and molds forensic scientists. How does a lab balance the need to tackle an increasing backlog with a diminishing budget while still finding time to mentor young forensic scientists? What do budding forensic scientists do when they discover the field is not sunglasses and Hummers[®]? Do they take matters into their own hands and become the next lab scandal? Or do they become disenchanting and leave the field entirely?

This presentation will examine the perceptions of what knowledge, skills, and abilities management expects of new hires before they start, the perspective of new hires and what they expect not only from the training program but from the job itself, and if there are ways to craft training programs to not only teach the required skills but also engage the full potential of the new hire. Using surveys sent to the management or section heads and the newest hires of forensic labs, this presentation will pinpoint areas of disconnect as well as mutual agreement. Questions will also be asked of educators as to what they feel their role is in molding these young minds. Possible solutions for resolving the dichotomy will be proposed.

Education, Mentor, Trainee

D72 Effect of Machine Laundering Additives on Human Blood

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After attending this presentation, attendees will know more about the interactions of laundry detergent and human blood.

This presentation will impact the forensic science community by helping investigators who find evidence in a washer determine the additive(s). They will also be better able to determine when to send out samples for DNA profiling, which in turn might save money for their department and provide a quicker turnaround time for labs since they would not have to handle unnecessary samples.

Perpetrators sometimes use household cleaners such as laundry detergents to try to hide or remove evidence at or from crime scenes. The effects of detergents containing active oxygen have been studied in the past but the effects of newer enzyme-based cleaners or green-based cleaners have not yet been examined. This research evaluates the effects of newer laundering additives on the detection and genotyping of DNA from human blood.

Two different treatments with detergent and human blood were evaluated for this study. To simulate the direct application of detergent onto a blood stain, one treatment entailed applying an equal volume (1.0mL) of detergent directly to human blood. A simulation of detergent concentrations encountered during laundering (using the detergent manufacturer's recommended concentrations) was also performed with heavily diluted detergent solutions. Eighteen different additives were tested including: detergents; detergent booster; pods; powder; pre-treatments; dryer sheets; and softeners. DNA was extracted from all samples using a phenol-chloroform protocol and isolated with an ethanol precipitation. A Real Time-Polymerase chain Reaction (RT-PCR) approach (Quantifiler[™]) was used to measure the amount of DNA recovered from each sample, with representative samples found to

contain DNA being genotyped.

Initial results indicate that the majority of the additives caused a greater loss of detectable human DNA when diluted rather than when undiluted. Detergents with borax, diethylene glycol, pentetic acid, Diethylene Triamine Pentaacetic Acid (DTPA), and disodium diaminostilbene disulfonate caused the greatest loss of DNA when diluted with water. Previous research has mentioned that active oxygen cleaners interfere with preliminary tests for human blood (such as luminol) but do allow DNA to be reliably genotyped. In these experiments, it was found that active oxygen detergents and/or boosters do significantly reduce quantities of DNA obtained from human blood to the point that DNA profiles may not be obtained after laundering with them. Laundry softener also resulted in a statistically significant reduction in the quantities of DNA obtained from these samples.

Further testing needs to be completed on the individual chemicals that are included in the laundry additives to determine which chemicals have the most effect on degrading the DNA. This research may identify a chemical to be removed from detergents to stop evidence from being destroyed. In addition, if investigators find evidence in a washer and can determine the additive(s), investigators may make a better decision as to when to send out samples for DNA profiling, which may save money for investigative agencies and result in quicker turnaround times for labs resulting from the handling of fewer samples.

Laundry, DNA, Persistence

D73 Why More Women Are Pursuing Forensic Science Graduate Degrees Instead of Other Science, Technology, Engineering, and Math (STEM) Degrees: A Qualitative Study

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After attending this presentation, attendees will evaluate reasons why women are pursuing forensic science graduate degrees. Attendees will also be able to assess reasons why women are discouraged from pursuing a Science, Technology, Engineering, and Math (STEM) degree at the undergraduate and graduate level of education.

This presentation will impact the forensic science community by examining the reasons why women are choosing to pursue forensic science graduate degrees over other STEM graduate degrees. The reasons why women are not choosing STEM graduate degrees can shape forensic science laboratory policies to prevent women from leaving the field of forensic science in the future.

Although women are qualified and capable of being successful in STEM majors, only a fifth to a third of the STEM fields are composed of women. The loss of talent, diversity, and creativity is cause for concern for the future. Forensic science undergraduate and graduate programs nationally average 78% of the student body as female. The purpose of this research was to determine why women are choosing forensic science as a career path over other STEM majors. A survey was sent to current female forensic science students and recent graduates. Based on their answers, many knew they wanted an undergraduate degree in forensic science before they started college. They were also greatly influenced by popular forensic television shows and fiction books.

The appeal of forensic science to the women surveyed is

mainly a combination of being able to do something different every day, something that has a direct application on people's lives, and can bring justice to a victim. These are things that are lost in many STEM research laboratories. The respondents stated that forensic science laboratory work is dictated by what crime has been committed and provides excitement and interest on a daily basis.

A follow-up research project tried to determine at what point during a woman's education she is mostly likely to be discouraged from pursuing a STEM undergraduate or graduate degree and what are the most common sources of that discouragement. The survey was sent to female university faculty teaching biology, chemistry, physics, biomedical sciences, forensic science, and medicine. Almost all the respondents decided to pursue a STEM degree prior to entering college. The types of discouragement for those pursuing STEM degrees identified in the survey were compared to the responses from women who received forensic science graduate degrees to identify any differences. Less than half of respondents were discouraged from pursuing a STEM degree either prior to or during their undergraduate degree primarily by family members. Those that chose forensic science as an undergraduate or graduate major were encouraged to pursue their degree.

Women, STEM Degree, Higher Education

D74 3D Facial Approximation: Lingering Problems and Improving Outcomes

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The goal of this presentation is to demonstrate the roles of both traditional and digital facial approximation (reconstruction) and to suggest appropriate applications for traditional, digital, and hybridized approaches to improve outcomes.

This presentation will impact the forensic science community by providing an evaluation of the current challenges inherent in facial approximation and pathways to overcome them. Organizations in need of this service should be able to discern which form of facial reconstruction is most useful to their efforts. While some agencies are better served by a completely digital or hybridized approximation, others might prefer a traditional approximation for display or educational purposes. Likewise, practitioners must examine and understand the rich complexity of skills necessary to perform facial approximations in both media in order to assess their strengths and weaknesses in the field.

The procedure as it currently exists suffers from several deficiencies. This is a field that requires an unusual blend of abilities: advanced sculpture; osteology; physical and forensic anthropology; and extraordinary observation skills in addition to knowledge of law enforcement procedures. Most people cannot perform well in all of these areas. Second, there are gaps in the science supporting the process of facial approximation. Tissue depth data is generally insufficient and whole racial or ethnic groups are under-represented. Further complicating matters, obesity and the growing number of individuals with multiracial ancestries is becoming more common, with limited tissue data existing for these societal trends. Lastly, the reliability of certain elements of the procedure can vary, with these differences often treated as equally important by practitioners. What are the most and least scientifically reliable elements to the reconstruction process? Can the least reliable elements be improved to make the results more predictable? These questions are addressed in the presentation.

Initially, learning the process of facial approximation in the traditional medium of clay is a valuable way to begin for several reasons: the highly specific challenges become immediately apparent; keying landmarks and interpreting the skull surface

with precision are better done in real space; and a developed tactile sense markedly improves digital sculpting. After training in traditional media, practitioners are more equipped to recognize and compensate for the deficiencies of digital approximation. Because traditional media is rarely used in law enforcement today, it becomes a challenge to maintain the strengths of traditional practice while integrating new media.

Although the advantages of digital media are obvious, it is just as clear that none of those advantages will overcome poor observational skills or a weak understanding of human osteology, deep, superficial, and surface structures, or the aging process. Design and rendering software builds on traditional fine arts skills and cannot mask their absence.

Unreliable results, high cost, and few practitioners are the reasons agencies do not use this technique more often. The expertise of artists is vital to the process, but not yet represented throughout the discipline. Interdisciplinary communication and cooperation would create more standardization and reliable results.

A key recommendation of this presentation is to seek improved communication and integration of the disciplines most directly linked to facial approximations, including law enforcement agencies, digital technicians, and traditionally trained artists. Many other disciplines may be consulted as determined by each case. Over-reliance on the technical side of digital approximations has led to mannequin-like reconstructions that look generic, thereby hindering the usefulness of the endeavor.

Conclusions on unreliable aspects of the process as well as best practices for the creation and the dissemination to the public will be detailed in the presentation. Additional recommendations regarding technical, scientific, communication, and dissemination concerns will be offered.

Facial Approximation, Facial Reconstruction, Forensic Art

D75 Management of Mass Disasters in Spain: The March 11th Madrid Train Bombing

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After attending this presentation, attendees will be familiar with the forensic investigation of victims of the terrorism attack that occurred on March 11, 2004, in Madrid, Spain, which was carried out by a collaboration of different forensic teams in Spain.

This presentation will impact the forensic science community by providing knowledge on the features of mass disaster management in Spain, highlighting the role of different forensic disciplines in the identification of the victims.

On March 11, 2004, during the morning rush hour between 7:39 a.m. and 7:42 a.m., ten improvised explosive devices were detonated on four commuter trains running from outside Madrid to the Atocha Railway Station, located near the center of the city. The ten explosions occurred almost simultaneously and resulted in 177 deaths, with more than 2,000 injured. There were subsequently 14 more in-hospital deaths (9 soon after admittance and 5 occurring later), bringing the ultimate death toll to 191. The 11-M Madrid commuter train bombings of 2004 constituted the second-largest terrorist attack to occur in Europe after the 1988 Lockerbie bombing, while the subsequent investigation became the most complex and wide-ranging forensic case in Spain's history.

The 11-M Madrid train bombings were a challenge for the teams' professionals involved in victim identification. Although

every mass disaster is unique, presenting a specific scenario, an accurate management of the situation is critical for proper victim identification. As a result, management, recovery, and laboratory methods of identification are all of great importance to an efficient resolution of those types of disasters.

A provisional morgue was established in the 6th pavilion of Institución Ferial de Madrid (IFEMA), where the different forensic teams worked on victim identification. Five work sites were created in the pavilion and the cadaver study was performed as follows: (1) Autopsy — three pathologists, three scientific police personnel, and a photographer worked together in this area where seven autopsy stations were located; (2) Radiology — two portable X-ray devices were taken from a nearby hospital. Two forensic pathologists and two radiology technicians performed the radiographic study of the victims; (3) DNA sample collection — a team of the National Institute of Toxicology took the required samples for DNA identification; and, (4) Forensic Anthropology and Forensic Odontology — two forensic odontologists, an anthropologist, and a pathology technician worked in the anthropological and odontological postmortem data collection of the victims.

Once the postmortem data was collected, a comparison with antemortem data was conducted, establishing positive identification of the decedent when possible.

The identification methods and percentage of identifications were as follows: fingerprints identified approximately 76% of the victims; dental, medical data, personal effects, tattoos, and family testimony were used to identify 8% of the victims; and DNA was used to identify 16% of the victims. Eighty percent of the victims were identified within 40 hours of the disaster.

In situations of mass disaster, the participation of the different forensic disciplines is needed to establish an identification protocol or to develop a new protocol in order to be prepared for future situations. For that reason, review of the management, recovery, and identification process developed during the 11-M Madrid train bombings may help to understand and improve the approach to these situations.

Terrorism, Bombing, Identification

D76 Assessing the Usefulness of Plant $\delta^{15}\text{N}$ as a New Forensic Tool to Identify Clandestine Graves

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After attending this presentation, attendees will understand a new, non-invasive forensic tool that may be useful in identifying clandestine graves.

This presentation will impact the forensic science community by demonstrating results from a pilot study indicating that new-growth grass samples collected above a burial and along a burial's edge have higher nitrogen isotope values than those collected off the grave.

Burials, including mass graves, are notoriously difficult to locate. As a result, forensic anthropologists and archaeologists often assist law enforcement teams searching for and recovering buried remains. Internationally, there is increasing interest in locating clandestine graves to aid in achieving accountability for mass atrocities and to further transitional justice processes. Homicide, genocide, and other crimes-against-humanity investigations are more challenging without a body, and victims' families often suffer

emotionally because of the lack of a body.^{1,2}

To potentially provide an additional tool to aid in the location of buried remains, an 18-month pilot study was conducted at the Forensic Anthropology Center at Texas State University, San Marcos, to test the hypothesis that plant $\delta^{15}\text{N}$ values could be used to identify clandestine graves.

The principle behind this study is based on the known differences in the ratio of the stable nitrogen isotopes in various biological organisms. Significant differences in $\delta^{15}\text{N}$ values exist between humans and naturally growing vegetation. Accordingly, a decomposing buried body will contribute nitrogen that is measurably ^{15}N -enriched as compared to most soil systems. The assimilation of this human-derived nitrogen should result in much higher $\delta^{15}\text{N}$ values in plants growing above bodies as compared to those that do not have access to this source of nitrogen.

To empirically test the principle and examine the usefulness of plant $\delta^{15}\text{N}$ as a forensic tool, a single cadaver was placed in a shallow grave, and the vegetation growing both above and beyond the grave was collected weekly from a defined grid. The basic questions addressed in this test were: (1) Do plants growing above interred bodies have distinct $\delta^{15}\text{N}$ values as compared to the surrounding vegetation?; (2) If so, when does this signature appear and how long does it persist after burial?; and, (3) How localized is the signature? Does it provide for high-resolution identification of graves or is the signature dispersed?

Isotopic measurements were obtained for a subset of the plant samples collected at 4, 15, 22, 24, 39, 61, 63, 71, 73, 77, 81, and 94 weeks post-burial. From weeks 61 onward, plants above the grave had $\delta^{15}\text{N}$ that were >20 per mil higher than those growing as little as 2m beyond the grave. Although a shift in $\delta^{15}\text{N}$ was anticipated, the differences were larger than expected and it is likely that soil fractionation processes may be further enhancing the ^{15}N -enrichment. While the experimental conditions were not ideal, the results still clearly indicate the effect buried bodies have on plant nitrogen isotopic values. More robust studies are now needed to explore the potential of the techniques as a useful forensic tool for locating clandestine graves.

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Clandestine Grave, Nitrogen Isotopes, Human Burials

D77 Lethal Railroad Injuries in Western Anatolia Eskisehir, Turkey: 2001-2010

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After attending this presentation, attendees will understand that transportation is not just a service that has a major social benefit but also a service that has both communal and individual costs. These costs are not just economical ones, but also include transportation duration, undesired environmental

effects, and accidents. The most important one among these costs is human deaths that occur due to accidents.¹⁻⁴ In Turkey, which has a total of 12,000km railroad web, only 5% of the load and passenger transportation is conducted via railroads. In Turkey, approximately 80 million individuals per year use the railroads as a mean of transportation. Seventy-five percent of these train rides are made by commuter trains.⁵

This presentation will impact the forensic science community by describing that in Turkey, where only 5% of all load and passenger transportation is being conducted via railways, it has been determined that railroad accidents and the deaths related to these accidents are higher than would be expected when compared to other countries in which the percentage of the railroad transportation exceeds that of Turkey. The results of this study show that security precautions in railroad stations, level crossings, and the railroad segments that are passing from residential areas should be increased. Railroad accidents are seldom compared to highway accidents but have much more lethal results.

Material and Method: In this study, 2,615 forensic-qualified deaths that occurred in Eskisehir between 2001 and 2010 were assessed. Thirty-eight of the 2,615 cases (1.5%) were railroad accidents and they have been reviewed.

Results: The ages of the cases varied between 7 and 72 years old (mean age: 37 (SD: 11.2)) and 81.6% were male. It was observed that there were injuries to the upper body (chest and back) in 78.9% of cases, to the abdomen and pelvis in 71.1% of cases, and injuries in both upper and lower extremities in 81.6% of the cases. Toxicological screening revealed ethyl alcohol presence in levels between 21mg/dL and 291mg/dL (average of 71mg/dL, SD: 43) in 16 (42%) of the cases.

After the examination of the autopsies and the judicial investigation, it was recorded that 76.3% of the cases were due to accident and 23.7% of the cases were suicide events. It was seen that 81.6% of the cases took place outside railroad stations and 52.6% occurred during summer months. Moreover, 57.9% of the incidents happened on either Friday, Saturday, or Sunday. Fifty-seven point nine percent lost their lives between 6:01 p.m. and 12:00 a.m.

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Railroad Accidents, Train Collision, Autopsy

D78 Taphonomically Important Fungi in Southern Illinois

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After attending this presentation, attendees will be aware of the variety of fungal communities (genus level) observed growing on decomposing human remains in southern Illinois, the importance of which is still being assessed. Knowledge of fungal presence

has been suggested as useful for estimates of postmortem interval, location of buried remains, identification of primary and secondary deposition sites, and suspect inclusion/exclusion in a criminal investigation.

This presentation will impact the forensic science community by demonstrating some of the types of fungus present in southern Illinois, which may be useful in estimation of postmortem interval, identifying post-deposition body movement, and season of deposition, among other forensically important aspects.

This presentation addresses the variety of fungal colonies observed growing on the decomposing human remains deposited at the Complex for Forensic Anthropology Research in southern Illinois. Fungal communities were observed growing on all human research subjects deposited on the ground surface (n=6), regardless of whether the individual mummified or progressed via a moist decomposition processes. Over the course of the decomposition process, a variety of colors of fungal communities were observed, including green, orange, white, pink, purple, red, yellow, and black. Fungal communities were observed as early as two days post-deposition and some colonies required more than two months to appear. This presentation reports on the genetic identification of those colonies present on a specific data collection date (March 27, 2013). Ongoing and future research will involve collection of an increased number of samples to identify the entire spectrum of forensically important fungi present in southern Illinois. Fungal samples were collected from representative colonies of fungi of each color present on the date of sampling. These samples were collected from the exposed skin surfaces of three different individuals. All three individuals were in the advanced stages of decomposition (TBS \geq 17).^{1,2} Samples included a green fungus from the right wrist, white fungus from the right forearm, and an orange fungus from the left hand of Research Subject 007 (RS007). Samples collected on Research Subject 008 (RS008) were of a green fungus on the right side of the upper chest and a white/pink fungus on the medial surface of the lower left leg at approximately midshaft. The lone sample from Research Subject 009 (RS009) was a white fungus collected from the left foot near the toes. Fungal samples were collected by scraping approximately 0.1g of material into an endomorph tube with approximately 0.5mL of 95% ethanol (EtOH) using a sterilized metal spatula and forceps. Six total samples were included.

Samples were dried, and DNA was extracted using a chelex bead extraction protocol. Following extraction, Polymerase Chain Reaction (PCR) was performed to amplify a region of the Internal Transcribed Spacer (ITS) gene using primers specific for fungi. Success of PCR was confirmed on an agarose gel stained with ethidium bromide. Successful samples were cleaned using exonuclease-shrimp alkaline phosphate (ExoSAP-IT[®]) and sequenced using an ABI[®] 3130xl Genetic Analyzer. Sequences were then annotated and concatenated in Sequencher[®] 5.10.

Of the six samples processed, four successfully yielded genetic sequences. The white fungal sample from RS007 did not amplify and the white fungal sample from RS009 did not yield a sufficient number of base pairs to successfully identify the fungus. The remaining four samples produced sequences of base pairs of varying lengths (442-568bp). The base pair sequences were processed using the Basic Local Alignment Search Tool (BLAST[®]) for nucleotide sequencing, which returned between one and five genus-level possible identifications for each sample. The genera identified include *Fusarium*, *Necria*, *Davidiella*, *Cladosporium*, *Sphaerulina*, *Mycophaearella*, *Gibberella*, and *Cordyceps*. The green fungus observed on the dorsal surface of the wrist of RS007 has been isolated as genus *Fusarium*. The other three samples are being assessed using a variety of ecological, biological, geographical, and climatological factors to narrow the results to a single genus for each sample. Further research is under way to

identify the taphonomic effects of these fungi and their potential for use in forensic investigations in southern Illinois.

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Taphonomy, Decomposition, Fungus

D79 Mentoring Forensics: An Explosive Application

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After attending this presentation, attendees will understand the key practices in the effective mentorship of foreign military forces in the area of bomb scene investigation, including goals and benefits associated with the employment and application of weapons intelligence transition teams.

This presentation will impact the forensic science community by highlighting an application of mentoring within forensics and the successful outcome of those efforts as well as presenting a challenge for forensic practitioners to identify opportunities to mentor individuals.

Hostile environments often involve a significant number of explosive events. After civilian populace and troop safety is insured, there is ample opportunity to conduct site exploitation due to real-world necessity and in the form of training and mentorship of friendly host nation forces. These tasks are primarily carried out by Weapon Intelligence Transition Teams (WITT). The mission of a WITT is to coach, train, and mentor host nation investigators in the law enforcement community at the local and federal levels. The goal is to create a system capable of investigating explosive incidents and completing the process through the judicial process to final disposition. WITTs support multiple command elements through operational, tactical, and administrative responsibilities for both coalition and host nation forces.

An effective WITT is comprised of approximately 25 individuals from several types of background and military branches. Coalition forces team members may include Explosive Ordnance Disposal (EOD), intelligence personnel, photographers, Law Enforcement Professional's (LEPs), interpreters, and a security element. Each of these individuals has the added task of providing mentorship to their host nation counterparts. Foreign national forces are often equal in size and strength with emphasis placed primarily on training and education of team members.

Weapons intelligence teams provide several forensic functions aimed at stabilizing the counterinsurgency environment. These functions include many of the efforts traditionally identified with the analysts of bomb scenes including: site security, evidence collection, location of epicenter, and documentation and reporting. The combat environment also demands additional investigative efforts including: forensic site exploitation, intelligence gathering, and Counter-Improvised Explosive Devices (C-IED). Forensic site exploitation allows analysis to identify several technical and tactical items from post-blast analysis. Intelligence information seeks to hinder enemy bomb-making capabilities through disruption of supply chains and bomb-maker activities. Intelligence information may include suppliers, evolution of insurgent Tactics, Techniques, and Procedures (TTP's), as well as emerging threats. Similarly, several pieces of information about the individual(s) involved can

be obtained and include training, region, affiliations, and other facilitators. C-IED activities seek to defeat enemy Improvised Explosive Devices (IEDs) through early detection or neutralization at the point of attack.

As with the individual team-member responsibilities described above, the host nation forces benefit from investigative efforts. Specifically, the aggressive use of forensic investigation aids Host Nation (HN) forces in securing the populace immediately after the investigation as well as long term after the withdrawal of coalition forces. This supports the stabilization of the counterinsurgency environment and has the additional goal of contributing to the creation of a functional judicial system, including the use of forensic capabilities, after the withdrawal of coalition forces.

The mission and goals of WITT ultimately result in numerous benefits in support of tactical and strategic mission accomplishment, each with the goal of gaining and improving forensic information for the purpose of timely and accurate dissemination of information. First, forensic and intelligence information obtained can be given to a forensic laboratory, such as the Joint Expeditionary Forensic Facility (JEFF) or the Joint IED Defeat Organization (JIEDDO) for additional, more-detailed analysis. The results of the analysis allow information to be collated from several tactical units and from multiple sources for wider dissemination throughout the Operating Environment (OE). The intelligence analyses provide information on enemy actions which often result in updated and improved Standard Operating Procedures (SOPs) by Coalition Forces (CF). These SOPs have a direct and measurable result on counter-IED capabilities and survivability. Similarly, the teams identify individual bomb makers' manufacture and geo-spatial patterns and trends which provide additional tools for coalition forces to avoid or prevent explosive attacks. Finally, WITT provides technical information encountered including timers, electrical components, initiators, containers, and shrapnel material on a myriad of explosives. Each of these efforts seeks to disrupt enemy efforts via enabling offensive operations and creating an environment not conducive to employing explosives. This is accomplished by the capture or termination of enemy bomb makers and increased force protection and local national security. The disruption of insurgent bomb-making capabilities can be readily observed in terms of number and complexity of explosive incidents and will ultimately decrease deaths for the civilian populace as well as host nation and coalition forces.

In conclusion, this presentation will provide an introduction to forensic capabilities of weapons intelligence teams and their efforts at exploitation of explosive incidents as well as creating long-term benefits for the host nation. Additionally, the many areas of forensic mentorship associated with WITTs will be explored despite the many challenges and obstacles faced.

Training, Mentorship, WITT

D80 Comparison and Assessment of Field Test Kits for Commonly Seized Drugs of Abuse

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After attending this presentation, attendees will learn the differences in various types of field test kits. The capability, advantages, and disadvantages of different field test kits to presumptively identify several compounds of interest, including marijuana, bath salts, and synthetic cannabinoids, will be demonstrated.

This presentation will impact the forensic science

community by providing an unbiased comparison and analysis of presumptive field test kits. This research will assess the strengths and weaknesses of three different field test kits available for testing marijuana. This research will also compare various field test kits that are available for presumptive testing of drugs that are commonly seen by law enforcement. Comparisons and assessments will include ease of use, number of compounds presumptively identified by a kit, and accuracy of identification. This presentation will allow law enforcement and the forensic community to make an educated decision about which field test kits best meet their needs for presumptive testing.

Field test kits have always been an integral part of law enforcement for the testing of commonly seized drugs. These kits must be simple to use and understand, while being robust and reliable. The basis of these presumptive test kits is the distinct color change associated with the addition of the compounds of interest. However, the quantity of sample introduced into the kit can adversely affect the color change of the resulting mixture and, therefore, the commonly used phrase "bigger is better" is not useful to this situation. For the marijuana test, officers sometimes utilize a large sample size which can cause issues with visualizing the color change and the excess material and broken glass can crowd the container, making the reagents and layers less visible. IDenta® Corporation has developed a different sampling system that could potentially eliminate these types of issues. This research will look at the different systems comparing IDenta® Corporation's system versus the traditional pouch test kit and the glass ampoule test kits.

The synthetic compounds are continuously changing with time and legislation. As changes to the molecules occur, law enforcement is still tasked with identifying which packages of "spice" are controlled or not controlled and the decision of whether or not to seize the package. Field test kits are sold with the promise of being able to help the officer identify synthetics. This research will look at test kits from different companies and identify kits that are easy to use and produce accurate results. Samples tested using field test kits will also be analyzed on a gas-chromatograph/mass spectrometer to identify which synthetic cannabinoids, if any, are present.

The last component of this research will look at using different field test kits to identify "bath salts." There are many different compounds that are sold as "bath salts," but is there a kit available that can identify them all using a color test or do officers need to carry around multiple kits? Issues that will be addressed include multiple kits versus one kit, officer safety, and ease of use.

Field Test Kits, Synthetic Cannabinoids, Bath Salts

D81 Blunt Force Trauma Injuries as Documented in Two Pennsylvania Society for the Prevention of Cruelty to Animals (SPCA) Humane Law Enforcement Animal Cruelty Cases

Susan C. Underkoffler, MFS, 350 E Erie Avenue, Philadelphia, PA 19134*

After attending this presentation, attendees will have a better understanding of the need for consistent and standardized utilization of forensic techniques in animal cruelty and neglect cases, the surprising findings thorough investigation can uncover, and the effects this has on prosecution. This presentation will also provide insights into the similarities between veterinarians and medical examiners and the methodologies they employ. Reinforcing the need for forensic science in non-human cases has the power to change laws, a necessity in combating not only animal

cruelty but crime in general, when one considers the correlation between animal cruelty and interpersonal violence.

This presentation will impact the forensic science community by serving not only as an example of forensic incorporation by the veterinary community but also the importance of thoroughness in all types of investigation and evidence collection, including necropsy. It will increase the general public's knowledge and awareness of anti-cruelty investigatory techniques typically underutilized or undervalued, and their resulting impacts in court. The findings also establish a starting point for further statistical analysis of successful court outcomes pertaining to animal cruelty cases.

This study is an attempt to demonstrate two humane law enforcement cases of blunt force trauma in canines resulting in death and what, if any, impact these documented injuries had on the case outcomes. In both cases, there were very few overt physical indicators of blunt force trauma injuries. It was only during necropsy that the injuries were recognizable; they included severe subcutaneous hemorrhage, liver lobe fracture, a ruptured renal capsule, intra-abdominal hemorrhage, blood aspiration, and skull fractures. The injuries and wounds were documented during and after discovery both in photographs and radiographs, and these visual aids were presented in court along with the officer's findings and other evidence. None of the injuries to either dog would have been physically obvious with a preliminary external exam which indicates the need for proper and complete forensic necropsies, including both internal and external exams in all cases of suspicious deaths to determine not only the cause but manner of death. A proper forensic necropsy should document all findings, even the lack of findings, along with their contributory or exculpatory nature.

The PSPCA encounters cases of animal cruelty on a continuing basis and has its own Humane Law Enforcement (HLE) division. It often works in conjunction with traditional law enforcement officers, animal control teams, and the veterinary community as a whole to combat animal crimes throughout the state, and often these cases are directly connected with other crimes, including domestic violence and drug abuse. PSPCA HLE officers investigated over 10,000 cases of animal cruelty in 2012.

Veterinary forensics is a new branch of forensic science that incorporates the applicability of science to animal laws. The use of forensic science in animal investigations is growing exponentially due to increased public awareness of the heinous nature of animal abuse cases and the resulting demand for stricter punishments for offenders. Evolving harsher laws and intolerance of animal crimes has prompted a greater need for forensic techniques in the investigations to aid in successful prosecutions, something formerly only seen in human cases. Necropsy is a powerful forensic tool that can often make or break a case.

Animal Cruelty, Trauma, Blunt Force

D82 Whistler Sled Dog Investigation: Exhumation Scene Mapping and Display of Canine Graves and Remains in British Columbia, Canada

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WITHDRAWN

Sled Dogs, Exhumation, Mapping

D83 Five Years of Fatal Dog Bites in Virginia: Case Studies

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After attending this presentation attendees will have a better understanding of the circumstances surrounding fatalities due to dog bites in Virginia.

This presentation will impact the forensic science community by providing case analyses of dog bite fatalities including discussion of the significant differences and similarities between the cases, in terms of injury pattern, dog breed, and victim age. Understanding these cases may prove useful in determining how to prevent fatal dog bites as well as which situations involving dogs can prove to have a fatal outcome.

Data: Seven cases of fatal dog bite cases occurred in Virginia in the last five years. The ages of the victims ranged from 6 months to 70-years-old with the average age being 38.8 years. There were two infants, ages 5 months and 1 year, but no children or adolescents. The remaining five cases were adults. The dogs were mostly medium- to large-sized dogs and most were pit bulls or pit bull-mix breeds. Most victims had punctures and lacerations to their faces and necks. Two out of the seven fatalities were female, four of the cases were White fatalities, there were two Hispanic fatalities, and one Black fatality.

Discussion: All of these attacks were unwitnessed and all attacks occurred within minutes of the times the victims were last known to be alive. Triggers for the attacks are unknown, except for two cases: in one case, the victim trained pit bulls and staged fights in his home; and, in the other case, the victim was moving toward the dog and startled it.

Injury patterns were similar, for the most part on the face and neck, except for one case where the victim was bitten on the leg and died from complications of the leg wound. Another case differed from the rest of the fatalities in that the victim, in addition to wounds on the head, had wounds to the arms and legs as well as defensive wounds to the hands.

The breeds of the dogs were mostly attack dogs, i.e., pit bulls and bulldogs. The exceptions were one case involving an Alaskan malamute and another case where the breed was not documented. They were all medium to large, stocky, and heavy breeds, and no small lap dogs were involved in the attacks. While investigating the cases, it was thought that most of the victims would be toddler-age children as opposed to adults. However, five out of the seven cases were adult fatalities, who presumably would be better able to protect themselves and/or not trigger or instigate dog attacks.

Conclusion: The majority of the injuries to the victims were to the head and neck, consistent with the areas dogs are likely to attack. The dogs involved in the fatalities were most often one of the “attack breeds:” pit bull, bulldog, and rottweilers. While there were a couple of infants involved, most victims were adults. None of the deaths involved children or adolescents (>1-year-old, <18-years-old). Further investigation will look into the similarities and differences of fatal dog attacks within a larger time frame.

Dog Attack, Fatalities, Virginia

D84 Using Geographic Information Systems (GIS) to Explore “Patterns or Practices” of Use of Excessive Force in Law Enforcement Agencies

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After attending this presentation, attendees will understand how spatial analyses can assist in determining the nature and extent of police use of force in a subject jurisdiction, as well as identifying the use of excessive force and any pattern or practice thereof.

This presentation will impact the forensic science community by providing a solid theoretical and empirical basis for pattern or practice investigations and by demonstrating how geospatial analysis can assist with monitoring, understanding, and responding to police use-of-force incidents. Experts involved in the analysis of police use of force for forensic purposes can expand their approach and increase the likelihood of producing meaningful and actionable results.

Descriptive statistical reports concerning the use of force by police typically tell a benign story: use of force is a relatively rare event, and when it is used, it tends to be proportional. When the data are considered in spatial context, the story often becomes less benign. Clustering of force incidents, particularly disproportional force incidents, may be more instructive in assessing the extent to which a pattern or practice of the use of excessive force exists.

Data for this presentation are drawn from more than 1,200 official use-of-force reports from the Seattle Police Department covering a recent two-and-one-quarter-year period. This period of time was associated with a Department of Justice investigation into alleged use of excessive force and racially biased policing in the city of Seattle. Levels of police force and levels of suspect resistance were classified based upon complementary scales following Alpert’s “force factor” method, and relevant Graham factors and other person and incident variables were coded. The data were geocoded to incident locations to enable spatial analyses. The focus of the presentation will be on the utility of weighted Kernel Density Estimation, with use-of-force incidents weighted by force factors.

Using these techniques, maps can be generated indicating concentrations (“hot spots”) of potentially disproportional use-of-force incidents, as opposed to concentrations of proportional force incidents. The underlying behaviors giving rise to police action can be contrasted across both weighted and unweighted use-of-force hot spots. Finally, concentrations of force incidents can be related, both visually and empirically, to other geospatial data, such as census demographics, area-located attitudinal survey data, and other data resources. A series of maps and spatial statistics will demonstrate how GIS can improve the quality of police monitoring and accountability mechanisms, particularly with regard to issues of social justice and questions about disparate impact in disadvantaged communities.

Police Use-of-Force, Pattern or Practice, GIS

D85 Forensic Analysis of Violence Against Women Murderers: Motives and Circumstances in Women Intimate Partner Violence Homicides and Their Consequences on Prevention

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After attending this presentation, attendees will better know some features and circumstances of the murderers in Violence Against Women (VAW). Usually, most of the papers and studies focus on victims, forgetting the aggressors and their motivations and circumstances to kill. The last World Health Organization (WHO) report on VAW (June 2013) established that 35% of women worldwide have experienced either physical and/or sexual intimate partner violence or non-partner sexual violence. This presentation also describes that perhaps as many as 38% of all murders of women are committed by an intimate partner after a history of violence and aggression. This means there is a similar percentage of men using this violence to gain their objectives.

This presentation will impact the forensic science community by explaining how to approach the experts to gain information as to the motives and circumstances related to these homicides and to obtain better knowledge about VAW homicides, resulting in collaboration to improve VAW risk assessment and to prevent these crimes.

An analysis of forensic reports and other elements included in the Spanish Judicial Sentences regarding the VAW homicides committed in Spain from 2001 to 2010 (n=309) (last data available) revealed a number of characteristics related to the aggressors and some unique case circumstances. Among them were: (1) percentage of cases where the aggressor was considered "guilty" by the courts (95.3%); (2) *modus operandi* and weapons used: the most frequent was "stabbing" (52.1%) and fire arms were used in 5.4% of the homicides; (3) homicides committed at home (76.7%); (4) time of day: most frequent was from 9:00 p.m. to 2:00 a.m. (36.9%); (5) aggressors with mental disorders (9.4%); (6) alcohol and drugs: the aggressor was considered under the effects of alcohol and/or drugs (6.9%); (7) victim and murderer were married in 43.2% of the cases; (8) previous violent episodes reported (previous complaint of VAW) (18.8%); (9) an existing protection order when the homicide was committed (11.3%); (10) women killed who had children (70.7%); and, (11) percentage of cases in which children were direct witnesses of the homicide (10.2%).

This and other data were analyzed to explain and further understand the circumstances surrounding VAW homicides. The results provide better knowledge of VAW homicides through the characteristics of the murderers and the circumstances related to the crime. In addition, the data also shows some features that challenge many myths and social beliefs typically associated to VAW and intimate partner homicides (i.e., that alcohol, drugs, or mental disorders are common among these criminals). The research identifies system weaknesses that may mask VAW cases and result in a failure to protect women and prevent serious aggressions and homicides. Finally, the results offer some suggestions in performing the forensic exam and risk assessment of these cases and in adopting measures to prevent future aggressions.

Violence Against Women, Murderers, Motives

D86 Serial Killer in Colombia: A Historical and Behavioral Perspective

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After attending this presentation, attendees will have a historical and behavioral analysis of serial killers in Colombia from the 20th century through the present day.

This presentation will impact the forensic science community by relating stories of the most notorious serial killers in Colombia, their victimology, *modus operandi*, and signature, plus the behavioral patterns they had in common despite the decades that separated them.

Colombia, a country in South America, has not avoided the phenomenon of serial killers that have been known as the most prolific killers, criminal history legends who crossed borders to leave their signature on crimes committed in neighboring countries such as Ecuador, Peru, and Brazil.

Dr. Mata, the Andes Strangler, the Charquito Sadist, the Beast, or the Cane Fields Monster are nicknames that produce fascination, but mostly awe and terror. The acts of these men have left a deep impact on hundreds of families over decades. Some of them currently represent a latent threat and cause constant fear in society because of their actions and the possibility that they may continue attacks after completing their sentences.

Nepomuceno Matallana was a bold and well-known fraud lawyer, who through charisma gained the trust of many people who were beat and tortured after being forced to sign documents that made Matallana the owner of their properties; he and his accomplices killed the victims and hid the bodies. Pedro Alonso Lopez, a humble man raised on a farm, ran away from home as a child to live homeless on the streets of Bogotá. He was jailed for theft, paid his fines, and was again on the streets. Lopez began his career as a serial killer of young girls in three countries in South America — his victims were his "dolls." Daniel Camargo Barbosa began his criminal career with the help of his wife by conning girls and then raping them. After being arrested for these crimes, Barbosa was sentenced to serve his sentence on Gorgona Island in the Colombian Pacific Sea. After escaping, he initiated an overland journey through several countries in which he committed more crimes against teenagers and young women. Perhaps the best known serial killer is Luis Alfredo Garavito Cubillos. He raped many children over several years without being caught, taking the lives of more than a hundred boys. Manuel Octavio Bermudez, hiding behind the facade of father and philanderer, was a brutal killer of children west of Colombia. Cases involving individuals with names like the Man Beast or the Mangones Monster (who managed to avoid law enforcement) have become urban legends because of the lack of case documentation.

Each of these killer's actions were unique; however, despite their differences and distinctions, research by the Criminal Behavior Analysis Group was concluded. The research focused on such items as the repetition of certain patterns, including how the selection of attack location and travel to other countries to commit crimes appears to have been determined by the geographical conditions of the Colombian territory, and how social factors may have provided access to victims. The findings of this research will be used to analyze active serial killer cases in Colombia.

Serial Killers, Criminal Behavior, Colombia

D87 Contribution of Postmortem Computed Tomography (CT) in Skeletal Trauma: About 28 Forensic Cases

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The goal of this presentation is to show the benefits and the limits of postmortem CT in skeletal damages.

This presentation will impact the forensic science community by permitting attendees to discuss the aims and the place of postmortem CT in modern forensic sciences.

Background: In France, postmortem full body CT is increasingly used for forensic purposes before a traditional autopsy is performed. However, the precise indications of CT are disputed in literature, ranging from almost never to the concept of virtopsy (i.e., in every case). Skeletal trauma is a well-known and accepted indication of CT scan. The goal of this preliminary study was to compare CT findings with autopsy results in forensic cases with severe trauma.

Method: In a group of 339 consecutive autopsies, 39 with a history of possible severe trauma had a postmortem CT before autopsy. In each case, lesions were classified according to the topography (skull, larynx, thorax, rachis, pelvis, upper and lower limbs). The radiologist and the forensic pathologist wrote independent reports which were compared by forensic pathologists who were not involved in these cases. Results between the two methods were compared with the McNemar's test.

Results: Twenty-one males and seven females, mean age-at-death was 33 years of age, and time elapsed between death and CT ranged from a few hours to five days.

Deaths resulted from traffic accidents (n=17), falls from height (n=8), assault and battery (n=1), and other causes (fall of a hydraulic hoist and a decomposed body (n=2)). CT provided a better description of skull fractures (especially the ones concerning basilar region and the calvarium) and seemed to have a better sensibility in identifying mandibular fractures (14 versus 9).

CT showed more non-comminuted scapular fractures and it allowed a more precise description of sacroiliac fractures. CT interest was even more obvious for detecting fractures of the extremities of long bones and hands. In upper limbs, 55 fractures were found using CT versus 29 in autopsy (p<0.001) and in lower limbs 84 fractures were found (CT) versus 36 (p<0.001) in autopsy.

CT could be more efficient for the diagnosis of fractures of dorsal vertebrae (22 vs. 16) whereas autopsy found more fractures of cervical vertebrae (21 vs. 15 (CT)). There were many discrepancies between both techniques in the description of fractures of the ribs, although the ones involving the posterior arch were seen better by autopsy.

Autopsy was much more efficient for detecting laryngo tracheal lesions (six on hyoid bone and five on the thyroid cartilage) as CT showed only two laryngeal fractures.

Discussion and Conclusion: More experience and training of the radiologist on postmortem material will probably improve the results of CT as well as the exchange of information with the forensic pathologist before releasing the report. Therefore, even if the small sample size does not allow definite conclusions,

it is thought that this study clearly supports the obligation of performing postmortem CT to assess skeletal damages in 2013.

However, CT missed some lesions of paramount forensic value such as laryngeal, hyoid, and posterior rib fractures which were detected by the autopsy.

At its best, skeletal trauma postmortem CT remains a complementary and valuable tool of forensic diagnosis. Getting rid of autopsy in any forensic case in 2013 is scientific nonsense and a judicial liability.

Autopsy, Computed Tomography, Comparison

D88 "One Giant Leap for Houston:" Shifting Paradigms for Forensic Services Operating Under the Umbrella of Police Management

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The goals of this presentation are to provide a history of the Houston Police Department Crime Lab and Identification Division and an overview of lessons learned, to explain the transformative progress, and to outline a novel model of forensic operation put into practice.

This presentation will impact the forensic science community by providing a set of clear and concise strategic steps that can be used in addressing the closure of a forensic unit, including how to regain the confidence of both a concerned public and the criminal justice system as a whole, and by setting in motion a novel model for administering a forensic operation based on the National Academy of Sciences (NAS) Report, *Strengthening Forensic Science in the United States: A Path Forward*.

Historically, the Houston Police Department (HPD) Crime Lab, like many other forensic operations across the nation, has had its share of forensic and economic challenges. After a series of investigative reports by a local TV station in 2002, the DNA unit closed its operation. Soon after, and with full cooperation from the lab and HPD, a full-scale review and assessment of the entire operation by an independent investigative team took place, lasting nearly two years, at a cost to Houston taxpayers well in excess of five million dollars. Several root causes in the historical crime lab were identified, from which a comprehensive final report and set of recommendations were produced. Four central themes emerged from these reports and systematic corrective action was taken by HPD.

Over the past ten years, there has been significant progress in the forensic operation while under the direction and control of the Houston Police Department. The City of Houston has undertaken significant expense and effort to transform the HPD Crime Lab and Identification Division. In 2005, the lab became accredited in Controlled Substances, Biology, Trace, Firearms, Questioned Documents, and Toxicology by the American Society of Crime Lab Directors-Laboratory Accreditation Board (ASCLD-LAB). In 2006, the DNA unit became fully operational and accredited. In 2012, the Latent Print facility was completely renovated into a state-of-the-art facility in preparation for ISO 17025 accreditation. Recently, units such as those housing crime scene and digital forensic investigators were brought under the umbrella of forensic services.

A comprehensive and rigorous quality assurance program was implemented. Quality of staffing has been dramatically improved with emphasis on experience, certifications and

educational credentials. Managers have been recruited with experience in laboratory management and expertise in forensic science. Rigorous training requirements have been imposed.

According to national reports, including the NAS Report, there is a need to remove public forensic labs from administrative control of law enforcement agencies or prosecutor's offices. An informal survey indicates that few public crime labs are outside the administrative control of law enforcement agencies or prosecutor's offices. In keeping pace with this recommendation, the city of Houston sought an innovative model of ensuring independence of their forensic operation from law enforcement and prosecutorial influence by establishing local government cooperation. This presentation will summarize the effort to make this vision to develop a model forensic operation a reality in Houston, Texas, and how it is structured with strong governance through a board of directors, technical advisory group, and an executive director. Finally, a summary of the progress made thus far will be presented, closing with the current development of a plan for a new forensic science center.

Houston, Independent Forensic Operation Model, NAS Report

D89 What is a Forensic Genealogist?

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The goal of this presentation is to provide an understanding of the principles of forensic genealogy with specific examples illustrating techniques and unusual resources used in forensic genealogy to establish kinship and identity.

This presentation will impact the forensic science community by the creation of an awareness of forensic genealogists as a resource for locating and investigating individuals in cases including those involving inheritance, the location of birth parents, and to serve as family references for DNA identification. Forensic genealogical techniques can be invaluable in providing new leads in cold cases. Attorneys and law enforcement will benefit from the knowledge gained concerning the capabilities of forensic genealogists.

Forensic genealogy deals with matters of kinship and identity. A forensic genealogist does not simply "look things up," but is often challenged to resolve identity issues with solutions that require insight and intuition. A forensic genealogist is consulted for his specialized knowledge of unusual sources of information. He may create a database pertinent to a case or may derive critical information by correlating data among multiple databases.

Some forensic genealogists are knowledgeable in the application of advanced DNA analysis techniques such as next generation sequencing or autosomal DNA testing. On a more basic level, when a law enforcement search for a DNA match in the Combined DNA Index System (CODIS) database proves unsuccessful, a forensic genealogist may be able to provide investigative leads by searching online genealogical databases for close or exact Y-DNA matches.

A private investigator is licensed to access personal data such as drivers' licenses and bank records that can produce an immediate identification. A forensic genealogist, on the other hand, establishes identity by correlating information that may be fragmented and that is often derived from obscure sources. While a private investigator may have numerous activities apart from personal identification tasks, a forensic genealogist specializes in identification. The scope of resources used by a forensic genealogist is much broader than that traditionally used by a private investigator. A private investigator is a general practitioner;

a forensic genealogist is a specialist.

International searches are challenging for more reasons than just the language barrier. Overseas resources can be scattered in foreign repositories, not easy to locate, and subject to unfamiliar regulations. Some of these difficulties are being reduced by social networking and the ability to communicate over email rather than on the phone. Online translators are essential.

This presentation will offer examples drawn from domestic and international case files to illustrate the creative methodology used by and the unusual resources available to forensic genealogists.

Forensic Genealogy, Identification, Kinship

D90 Automated Identification Technologies: Improving the Management of Forensic Evidence

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After attending this presentation, attendees will learn about Automated Identification Technologies (AIT), such as bar coding and Radio Frequency Identification (RFID), and an assessment conducted by the Technical Working Group on Biological Evidence to evaluate AIT's usefulness in forensic evidence management.

This presentation will impact the forensic science community by providing insight into new technologies that can aid in the management, tracking, and preservation of evidence.

Forensic science laboratories and law enforcement agencies have increasingly used AIT such as bar coding and RFID to track and manage assets such as forensic evidence, firearms, and personnel. In August 2010, the National Institute of Standards and Technology (NIST) and the National Institute of Justice (NIJ) convened the *Technical Working Group on Biological Evidence Preservation* "to create best practices and guidance to ensure the integrity, prevent the loss, and reduce the premature destruction of biological evidence after collection through post-conviction proceedings." While the best practices handbook was being produced, an assessment of automated identification technologies was conducted to evaluate its usefulness in improving the management of forensic evidence. As a result of the assessment, the group created recommendation IV-6 of *The Biological Evidence Preservation Handbook* which reads, "Overall, it is highly recommended that jurisdictions consider automated identification technologies to enhance chain-of-custody recordkeeping and tracking, to facilitate inventories, and to allow for efficient retrieval of evidence."

This presentation will include discussion of a report summarizing the findings of the AIT assessment. The report includes insights into the barriers facing the adoption of AIT within the law enforcement industry, the current business processes that can be benefited by injection of AIT, and a Return On Investment (ROI) analysis of RFID's use in property and evidence rooms. Finally, this report will address the foundational principles that must be established in order to achieve industry-wide adoption and the strategic next steps that should be undertaken by local law enforcement agencies and high-level federal stakeholders.

Evidence, RFID, NIST

D91 Manners of Death in Perpetrators of Domestic Violence

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The goals of this presentation are to: (1) observe different relationships of domestic violence; (2) review findings of death investigations; (3) compare findings at scene with those at autopsy; (4) discuss various manners of death associated with the perpetrators of domestic violence; and, (5) determine recurring patterns of fatality and harm that arise from domestic violence.

This presentation will impact the forensic science community by discussing how professionals involved in the investigation and management of domestic violence can be aware of the substantial risk of harm, including death, that a perpetrator does not expect but may face as a result of his/her acts. The information collected at the death scene, including photography, witness statements, prior domestic violence history, and the written narrative by the death investigator, will be important information later to Domestic Violence Review Teams in identifying risk factors and implementing intervention and prevention programs.

Hypothesis: Domestic violence can have fatal consequences to the perpetrator as well as the victim, resulting in three different manners of death including homicide, suicide, and accident.

Content and Methods: Cases from the Jackson County Medical Examiner's Office during 2009-2011 were reviewed. Investigator reports that indicated a history of domestic violence were analyzed. The consequences of domestic violence on the perpetrators were sought, particularly regarding injury or death that may have been related to the incident that brought the domestic situation to the medical examiner's attention. Four cases were found in which the perpetrator became a fatality as a result of active or recent domestic violence.

Results: One case was a classic homicide-suicide, in which the husband perpetrator killed his victim wife, and then shot himself. A second case was a suicide, in which the wife perpetrator hanged herself in order to avoid a court date with anticipated consequences for her previous acts. Third was a case in which the manner of death was an accident, where the long-term male companion perpetrator died from excited delirium syndrome related to heavy alcohol intake and vigorous resistance to police detention when they responded to a call for help because of active domestic violence from his victim girlfriend. The fourth and final case occurred over several days and involved three homicide victims and eventually ended with the perpetrator being shot by police. The male subject had killed one woman, a suspected drug-dealer companion of his, before returning home and, for some unknown reason, killing his sleeping wife and goddaughter. When confronted by police, the perpetrator fired shots at the police, who returned fire. The manner was ruled as homicide.

Conclusion: Domestic violence can be as lethal to the perpetrator as the intended victim. The medical examiner has a substantial role in investigating, documenting, and reporting these fatalities. Circumstances resulting in death of the perpetrator may include homicide, suicide, and accident as manners of death.

Forensic Pathology, Forensic Science, Violence

D92 Teaching Forensic Science as a Science and as a Profession

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After attending this presentation, attendees will understand the problem of conveying to students the features of forensic science that distinguish forensic science from other scientific disciplines. Attendees will also understand how professional ethics and quality management concepts can be introduced to neophyte forensic scientists through lectures, readings, and writing assignments.

This presentation will impact the forensic science community by demonstrating how beginning forensic scientists come to understand forensic science as a historical science, to understand the importance of professional ethics, and to understand the vital role of laboratory quality management systems.

Graduate students entering master's degree programs in forensic science bring with them a number of misconceptions. Their understanding of forensic science has largely been informed by television programs such as *CSI* and *Bones*, in which crime scene investigators collect evidence, perform laboratory analyses, and interview and interrogate witnesses and suspects. Moreover, most undergraduate students in the natural sciences have been educated in experimental science, rather than historical science. In experimental science, researchers form hypotheses which predict the outcome of future experiments; in historical science, researchers look for traces of past events and try to reconstruct those events. Undergraduate science students (unless they have had occasion to work in an industrial laboratory) will not have had significant exposure to the concept of a laboratory quality management system (requiring, for example, standardized laboratory procedures which are ubiquitous in forensic science laboratories). Finally, entering graduate students will have little concept of professional ethics, in forensic science or in any other scientific discipline.

These lacunae in students' knowledge of forensic science are addressed in an introductory graduate seminar course required of all Master of Forensic Science students, regardless of their area of concentration. This course combines talks by departmental faculty and forensic science practitioners on a variety of topics, including scientific method, professional conduct, professional ethics, and quality assurance/quality control. These talks are supplemented by in-class discussions, readings, and writing exercises. For example, students read articles on the distinction between experimental and historical sciences and then write a "Logic of ..." paper which follows the rubric developed by Doctors Richard Paul and Linda Elder for the Foundation for Critical Thinking. In this exercise, students answer the following questions about forensic science in general or a selected forensic science sub-discipline (e.g., forensic molecular biology, trace evidence analysis, or forensic toxicology):

- What is the purpose of this field?
- What question(s) does the field seek to answer?
- What sort of information does the field use to answer questions?
- What interpretations of data do practitioners in this field make? What inferences do they draw?
- What concepts underlie this field?
- What are the key assumptions underlying this field?
- What are the societal implications and consequences of work in this field?
- What is the intellectual point of view of practitioners of this field?

In-class discussions of professional ethics are supplemented by readings in a leading textbook on professional ethics in forensic science and by the codes of ethics of a number of forensic science professional organizations. Student writing

assignments ask for responses to a variety of ethical dilemmas that might arise in the course of a forensic science career; students must cite the relevant professional codes of ethics in support of their resolutions of the ethical dilemmas. To drive home the need for quality management systems in forensic science laboratories (and also the need for codes of professional ethics), students read newspaper reports and other documents relating to the cases of Fred Zain and Annie Dookhan. The students complete a writing assignment in which they are required to analyze these cases and discuss the ways in which the laboratories' quality management systems were compromised. Students were then asked to suggest changes in laboratory policies that could prevent such episodes in the future.

Scientific Method, Ethics, Quality Management

D93 Deaths Due to Excited Delirium in Psychiatric Patients and Unassociated With Stimulants

Theresa G. DiMaio, BSN, 10 Carriage Hills, San Antonio, TX 78257; and Vincent J.M. Di Maio, MD, 10 Carriage Hills, San Antonio, TX 78257*

After attending this presentation, attendees will know that deaths due to excited delirium can occur in psychiatric patients unassociated with stimulants such as cocaine. The mechanism of death, a cardiac arrhythmia induced by abnormally high levels of catecholamines due to the mental illness itself and to the struggle, will be discussed as will the inability to resuscitate these individuals due to the nature of the arrhythmia.

This presentation will impact the forensic science community by showing that deaths from excited delirium do not have to be associated with the use of stimulants, have the same presentation as cases due to stimulants, and therefore can be prevented by the same treatment methods used with stimulant cases.

Delirium involves an acute (minutes to hours), transient disturbance in consciousness and cognition. There is disorientation; disorganized and inconsistent thought processes; inability to distinguish reality from hallucinations; disturbances in speech; disorientation to time and place; and misidentification of individuals. When the delirium involves combative and/or violent behavior, it is termed Excited Delirium (ED). Excited Delirium Syndrome (EDS) involves the sudden death of an individual, during or following an episode of excited delirium, in which an autopsy fails to reveal evidence of sufficient trauma or natural disease to explain the death. Individuals in ED are often resistant to the effects of pepper spray or application of an electro-muscular disruption device (e.g., TASER®).

Deaths due to EDS were originally described in individuals with intrinsic mental illness, specifically schizophrenia and/or bipolar disease. In our present society, they are more commonly associated with abuse of illegal stimulants such as cocaine or methamphetamine. Because of this, many individuals mistakenly believe that EDS only occurs in association with drugs. In fact, a significant number of individuals dying of EDS are individuals with intrinsic mental disease with drugs playing no role in the death.

The mechanism of death in cases of EDS appears to be a cardiac arrhythmia induced by abnormally high levels of catecholamines (epinephrine and norepinephrine) due to the intrinsic mental disease or illegal stimulants combined with elevated levels due to the struggle. In the case of individuals using illegal stimulants, the elevated levels of catecholamines are associated with the physiological action of these drugs. In addition, these

drugs also have a direct cardio-toxic action on cardiac muscle.

Individuals with intrinsic mental disease, such as schizophrenia, quite frequently experience episodes of acute psychosis/excited delirium. This can be due to failure to take medications, use of drugs of abuse, perceptions of a hostile environment, or a perceived provocation. This tendency to develop excited delirium in schizophrenics is aggravated by the fact that in schizophrenia, there is a disturbance in the metabolism of norepinephrine (NE) both in the brain and peripherally. Numerous studies in both medicated and unmedicated patients with schizophrenia have found elevated NE concentrations in samples obtained from plasma, cerebrospinal fluid, and postmortem brain tissue. In addition, stress in individuals with schizophrenia results in reactive blood levels of NE higher than reactive levels in normal individuals.

Individuals with EDS going into cardiac arrest are rarely resuscitated successfully. The most likely explanation for this is the nature of the arrhythmia. Fatal arrhythmias fall into two general categories: tachy-arrhythmias such as ventricular fibrillation and brady-arrhythmia-asystole. The latter goes from bradycardia to Pulseless Electrical Activity (PEA) to asystole. While successful resuscitation rates as high as 59% have been reported with tachy-arrhythmias due to cardiac disease, with brady-arrhythmia-asystole, successful resuscitation is measured in the low single digits at best. Thus, in a prospective study of 148 subjects who went into cardiac arrest in casinos, 105 subjects had an initial cardiac rhythm of ventricular fibrillation, 17 had PEA, and 26 had asystole. Of the subjects whose initial cardiac rhythm was not ventricular fibrillation, none survived to be discharged from the hospital. Automated external defibrillators were used on the 105 patients with ventricular fibrillation. In 90 instances where the collapse was observed, 53 (59%) survived to be discharged from the hospital. Three of the 15 whose collapse was not observed survived.

This study presents 11 cases of EDS, 7 due to psychosis, where EMTs were present at the scene of a cardiac arrest and immediately documented the arrhythmia. Of the 11 cases: six showed asystole; one showed PEA; two had bradycardia; one was a progression from a normal sinus rhythm to bradycardia, to PEA, and then asystole; and, one case where an EKG showed borderline bradycardia with a second EKG a few minutes later showing asystole. This research suggests that this last case should be classified as a brady-arrhythmia progressing to asystole. The nature of the arrhythmias observed in these cases of EDS may explain the virtual absence of successful resuscitation from this entity.

Excited Delirium, Psychiatric Deaths, Catecholamines

D94 Death by Starvation: A Case of Child Neglect

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After attending this presentation, attendees will understand the difference between marasmus, malnutrition, and failure to thrive and how these diagnoses may impact a child death investigation.

This presentation will impact the forensic science community by providing information regarding fatal child starvation, the death investigation surrounding one case, and the legal outcomes which resulted.

Fatal child starvation is uncommon in the United States and much can be learned from those cases in which it does occur. This presentation describes a case study of an infant who died at nine months of age who weighed less than his birth weight. This case study will highlight the importance of a team approach

while investigating deaths of infants and children. The case study will include information regarding the interview techniques, the scene investigation, the 911 recording, records review, autopsy, toxicology, genetic testing, and the child death review which were all utilized prior to ruling the death a homicide due to child neglect.

This case encountered legal challenges due to disagreeing opinions regarding the cause of the child's "failure to thrive." This case will highlight how the mention of a potential undiagnosed genetic disorder may complicate an investigation. Following autopsy, it was suggested that the infant may suffer from a genetic disorder, DiGeorge Syndrome. As a result of that suggestion, it was important to conduct a full genetic screen to determine if this child suffered from such a syndrome and if that syndrome played a role in the child's death.

Discussion will include the common physical findings and characteristics exhibited in pediatric starvation, the differences between marasmus, malnutrition, and failure to thrive, as well as the importance of the death scene investigation and medical record review. This case study is a useful tool in explaining the importance of an immediate death scene investigation and the information that may be obtained. This case also highlights the importance of obtaining and reviewing all medical, social, educational, and criminal records as a part of a complete child death investigation and how those records may be used in determining the manner of death and potential criminal charges which may result.

This case also highlights the importance of having qualified specialists conduct forensic interviews on living children who may have witnessed/participated in the care of a sibling who died. Information revealed during child forensic interviews, in this case, were found to provide very important information regarding the care and/or neglect which occurred, who the primary care takers were in the home, and in recommending counseling and treatment for those living children.

Child Death Investigation, Starvation, Marasmus

D95 Forensic Microscopy for an Undergraduate Student

Gina Londino, MS, 402 N Blackford Street, LD 326, Indianapolis, IN 46202*

After attending this presentation, attendees will learn how different types of microscopy are taught in an undergraduate classroom. Specific laboratory exercises that focus on four types of microscopy will be discussed with the goal of understanding commonly used microscopes in a forensic laboratory to include stereomicroscopy, compound light microscopy, polarizing light microscopy, and instrumental microscopy.

This presentation will impact the forensic science community by showing specific microscopy exercises being developed and offered to students in undergraduate forensic science programs and courses.

In a growing forensic science program, there is a need to offer courses which specifically teach approaches to individual forensic techniques used repeatedly in most forensic science courses. There are courses specific to forensic chemistry, which may also be broken into illicit drug analysis, explosives, or composition of ink. In forensic biology courses, there are separate courses on genetics, DNA, and statistics. Universities that offer forensic science courses also focus on impression evidence, offering an entire course solely on fingerprint and footwear impressions. Photography and crime scene investigation are usually individual courses as well. One scientific instrument that is common across nearly all forensic courses is the microscope.

Microscopes are used to examine multiple types of

evidence from chemistry, biology, impressions, and crime scene. However, microscopy is usually not taught as a stand-alone course even though the use of microscopy is seen over and over again. Microscopy should also be taught as an individual course similar to other forensic courses. This type of course would allow students to focus on proper microscope use and application to the analysis of different types of evidence. This application can then be used for a multitude of forensic sciences.

Each type of microscope can be the course focus and then different types of evidence can be examined using the techniques discussed. Students will be able to understand how each type of microscope is different and offers different abilities to the forensic scientist. All types of evidence can be examined using microscopy; therefore, this course is not limited to the forensic chemist, biologist, etc. The approach taken in this course teaches students how to handle stereomicroscopes, compound light microscopes, polarizing light microscopes, and instrumental microscopes. Each type of microscope will be broken down into components, teaching students how the microscope functions and the advantages and disadvantages to each type of microscope.

Once students have a general knowledge about each microscopy technique, a variety of evidence is used to emphasize how each microscope is utilized in a forensic laboratory. Due to advances in forensic science program curriculum, there has been a need seen for a course that covers just forensic microscopy. Forensic science education should consist of basic forensic science courses; however, more advanced coursework should be included with laboratory components.

During this presentation, discussion will consist of the development, implantation, and modifications made to a forensic microscopy course in a Forensic Science Education Programs Accreditation Commission (FEPAC) accredited forensic science program. This will cover the course curriculum, types of students in the course, and changes made within the course.

Forensic Education, Microscopy,



Jurisprudence



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E1 Forensic DNA Phenotyping (FDP) From an Ethico-Legal Perspective

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After attending this presentation, attendees will be able to understand the difference between traditional forensic DNA typing and Forensic DNA Phenotyping (FDP), the current importance of the latter, and the main legal and ethical issues involved in its use in forensic sciences.

This presentation will impact the forensic science community by highlighting the legal and ethical issues concerning FDP, since it is a new and largely unregulated area of modern criminal investigation.

In criminal investigations, traditional DNA typing determines whether two samples (one from the crime scene) are from the same person. But since ethical, legal, and economic problems prevent creation of a universal DNA profile database in the majority of countries, this technology is of little use if police don't have a suspect. On the other hand FDP, using the DNA left at a crime scene, has the possibility of creating a genetically based description of donor appearance that police can use to find a suspect.

Currently, FDP relies on commercially available tools that produce information about eye/hair color (directly DNA-linked characteristics) or biogeographic ancestry allowing inference of characteristics like skin/hair color, stature and facial structure. It is also possible to use large databases to determine a male's surname based on DNA analysis. Since efforts to unlock the deeper secrets of the human genome continue, it is expected that DNA research may reveal further correlations between genes and traits, including propensity for certain types of disease/behavior. In fact, a phenotypic trait is not affected by only one gene. The expression process relies on interaction with other genes and external factors. Thus, FDP can potentially reveal a significant and expanding amount of information that invokes genetic privacy concerns related to what information can be obtained/disclosed. In this context, the donor's "right not to know" should not be neglected. There are also concerns about the possibility of using FDP to infer "bad genotypes" related to crime or antisocial behavior. Furthermore, since ethnic profiling is one of the most prominent applications of phenotyping, it can encourage stigmatization/discrimination. But genetic ancestry does not equal race. Race is a concept influenced by biologic and sociocultural patterns, is not well defined and does not necessarily predict someone's appearance. Still, the significant level of the uncertainty of FDP predictions, when compared to traditional DNA typing, complicates the interpretation of the results (e.g., by police).

It is important that justice systems anticipate and react to the problems raised by this developing method but, truth be

told, most of the pointed concerns are speculative or hypothetical. People are worried about what they do not know or what might happen. Although some concrete instances of abuse already happened it is difficult to conclude whether they represent the tip of the iceberg or are simply outliers. Most countries, like Portugal, have not yet taken a side on this matter. Nevertheless, people should not mistake such silence for an implicit prohibition of FDP. In the United Kingdom, for instance, phenotyping for ethnic origin and hair color is regularly done, and many U.S. states' legislation (e.g., Texas) suggests that it is allowed as well. According to current knowledge, Netherlands is the only country that has legislation that explicitly allows the use of FDP to determine biogeographic ancestry and specific externally perceptible traits (visible from birth and unrelated to disease), provided that the technology used is sufficiently reliable. The U.S. states of Vermont and Utah only exclude the determination of genetic diseases. On the other hand, Belgium, Germany, and the U.S. states of Indiana, Rhode Island, and Wyoming explicitly prohibit deriving physical traits, other than gender, from DNA.

This ongoing process is important because it redefines the rights and status of the suspect's biological sample and its credibility as criminal evidence. But it should be noted that circumstances of specific criminal investigations vary, as does the reason why and what phenotypic information is relevant. It may be necessary to allow or restrain the use of particular visible or non-visible traits in specific cases. Legislation should not be framed in terms of prohibiting the adoption of developments in DNA technology. Efficient policing can prevent prosecutors from failing to obtain a conviction due to key evidence being ruled inadmissible and defendants from being subject to unnecessary privacy breaches.

Forensic Phenotyping, DNA, Ethical Issues

E2 The Amanda Knox Case: A New Scenario After the Supreme Court Verdict

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This goal of this presentation is to expose the reasons for which the Supreme Court set aside the acquittal handed down by the Court of Appeal of Perugia, ordering a new trial, with particular reference to: (1) the appointment of the board of experts in the second degree (and the subsequent declaration of inoperability of genetic tests made by prosecution); and, (2) the possibility, according to the prosecution, of examining a new Low Template (LT)-DNA evidence on the knife presumed to be the murder weapon, although it was never tested during the appeal trial.

This presentation will impact the forensic science community by underlining the procedural difficulty of demonstrating the possible contamination of genetic traces (not recognized sufficient, for the Italian Supreme Court, for the demonstration of the violation of analytical protocols). It will also highlight the procedural difficulties for the judiciary of overcoming the scientific theories of the opposing parties.

After attending this presentation, attendees will be able to understand the scientific and judicial issues that have characterized the criminal trial held in Italy against Amanda Knox who was accused of murder.

This case refers to the murder of Meredith Kercher, a young British student, who was found dead in her apartment in Perugia on November 2, 2007. According to the prosecutor's investigation, the murder was committed by Amanda Knox, Raffaele Sollecito, and Rudy Guede. Knox and Sollecito were indicted on murder charges on October 28, 2008. Guede was found guilty of murder in his fast-track trial and sentenced to 30 years.

The evidence offered by the prosecution was circumstantial. No witnesses were present at the time of the crime. Among other evidence, the key to the guilt of Knox and Sollecito was mainly represented by the scientific investigation of two findings: a hook from the victim's bra and a knife found at Raffaele's house. The first of these two elements (the hook from the bra) was found during the first crime search made by the police the day after the murder. But it inexplicably gets lost and is found and collected 46 days after the crime, during new technical activities at the crime site. According to the prosecution theory based on the DNA analyses made by the forensic science lab, the hook shows genetic material that matches Sollecito's, indicating his presence at the crime scene during the murder. According to the defense theory, the DNA found on the hook is due to contamination occurring from technical activities made at the crime scene during those 46 days.

The second element, the knife, showed DNA on the blade that matched the victim and on the handle that matched Amanda. According to the prosecution theory, this was the "murder weapon" and Amanda was the person who stabbed Meredith. This item has clear circumstantial evidence weight. According to the defense theory, the DNA found on the blade is LCN-DNA, thus too low in quantity to support a reliable conclusion, especially if it is to be linked to Amanda's guilt.

The court of first degree accepted the experts' assessments of the prosecution about the collection, preservation, and analyses of scientific evidence. Contrary to normal practice Italian courts, which usually appoint an independent expert to settle disputes among the experts, the Court rejects the request made by the defense.

After eleven months of trial, the court found Knox and Sollecito guilty on all counts in the stabbing death of Meredith Kercher. Sollecito received a 25-year sentence; Knox 26-years. On November 24, 2010, Knox and Sollecito's murder appeal process began.

Forensic specialists appointed by the second degree court told the court that DNA evidence linking Knox to the alleged murder weapon was unsound; while they agreed Knox's DNA was present on the knife handle, tests for Kercher's DNA were unreliable. The sample; however, was so small that forensic scientists were not able to double-test it in accordance with international forensic science rules, which Knox's legal team said raised doubts about its validity.

Court-appointed experts testified that police forensic scientists involved in the murder case made a series of glaring errors during their investigation. In a point-by-point deconstruction, the experts said that because of the errors made by police during the original investigation, the evidence against Knox and Sollecito should be considered "inadmissible." On October 3, 2011, an Italian jury overturned the 2009 murder conviction of Amanda Knox and Raffaele Sollecito.

The attorney general appealed all decisions of the Court of Appeal, asking the Supreme Court to set aside the acquittal. On March 26, 2013, the Supreme Court censored the verdict of second degree in many respects, including the misjudgment of the circumstantial evidence (which is only hinted at in this presentation).

The Supreme Court also ruled that the Court of Appeal

did not adequately justify its decision to appoint a Board of Experts in order to resolve the dispute between the parties; that it is for the defense to prove the possible contamination of the samples, it not being sufficient to prove the non-compliance of protocols for the collection and the analysis of evidence; and that it is necessary to examine a third sample found on the blade of the knife, assumed to be the murder weapon, although it is an LT-DNA sample (which the experts had considered unnecessary to analyze).

Amanda Knox, LT DNA, Italian Supreme Court

E3 The Use and Misuse of Inquisitorial Experts

Joelle Vuille, PhD, University of California, Social Ecology II, Irvine, CA 92697; and William C. Thompson, PhD, JD, University of California, Dept of Criminology, Law & Society, Irvine, CA 92697*

After attending this presentation, attendees will understand how inquisitorial jurisdictions differ from adversarial systems in their use of scientific experts and what the advantages and shortcomings of each system are.

This presentation will impact the forensic science community by encouraging them to rethink some stereotypes that may have been held in the context of comparative criminal procedure in general and the use of court-appointed experts in particular.

Court-appointed experts are often depicted as a miracle solution in the legal and scientific literature: being supposedly neutral, they deliver high-quality scientific evidence in a non-partisan way. But are experts the only variable to consider when discussing the relative advantages and shortcomings of adversarial and inquisitorial experts? Could there be other mechanisms influencing the overall quality of scientific expertise in a given criminal justice system?

An empirical study was conducted comparing the use of scientific evidence in the United States and in Switzerland, a typical inquisitorial jurisdiction. Semi-structured interviews were conducted with defense attorneys in both jurisdictions to assess their attitudes toward scientific evidence and to collect data on their experiences in this context. Since almost all Swiss experts being court-appointed, it was expected that many of the problems encountered in adversarial jurisdictions (such as bias, extreme polarization, misunderstanding of expert conclusions by fact-finders) would be non-existent. The results of the study showed that the situation is more nuanced and complicated than usually described in academic circles, and that, if inquisitorial jurisdictions avoid certain pitfalls typical of adversarial systems, they also create problems of their own.

For instance, contrary to expectations, lack of transparency was denounced by all Swiss interviewees as being a prominent feature of the inquisitorial use of experts. Swiss attorneys also showed a high level of trust in scientific experts; yet, this trust appeared to be uninformed. Their knowledge of (and interest in) scientific evidence seemed very limited, and their appraisal of the probative weight of the evidence appeared to be based largely on cognitive heuristics. This led to a passive behavior that even the personal experience of scientific errors could not rouse. On the contrary, skepticism was a characteristic of all American interviewees, who appeared highly motivated to scrutinize the work carried out by prosecution experts.

Such a difference in attitudes between Swiss and American attorneys cannot be totally justified by objective differences in the quality of the scientific evidence to which they are confronted. Consequently, traditional legal scholarship praising the advantages of inquisitorial over adversarial experts should be considered with

care. It might well be that there is an epistemic advantage in the use of adversarial experts.

Inquisitorial, Adversarial, DNA Evidence

E4 Helping Juries and Officers of the Courts Make Sense of Statistics in Forensic Science: Update From the Working Group on Presenting Forensic Science Evidence Using Quantitative and Qualitative Terms (QQWG)

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After attending this presentation, attendees will learn about the nature of forensic inferences, the psychology of effective communication, and best practices related to presenting statistical information in testimony and reports.

This presentation will impact the forensic science community by informing forensic science practitioners, managers, and other key criminal justice system stakeholders how statistics can be effectively communicated in testimony and written reports. This presentation will provide better understanding of how numeracy, the ability to process basic probabilities and numerical concepts, affects how one processes qualitative and quantitative data.

The National Institute of Standards and Technology has partnered with the Pennsylvania State University to establish a working group that will propose generally applicable best practices for reporting relevant statistical information about forensic evidence (e.g., quantitative measurements, expressions of uncertainty or error probabilities in measurements or conclusions, and validation studies). To meet its charge, the working group reviewed studies related to the presentation of statistical information to lay individuals that have been published in forensic, legal, social science, and statistical literature. The group also reviewed how forensic science evidence and other types of expert or scientific evidence (e.g., medical evidence) have been presented in court in the United States (and in the similar legal system in the United Kingdom). This presentation will provide the findings and recommendations of the group.

Qualitative/Quantitative Data, Numeracy, Testimony

E5 Emergence of Scientific Latent Print Practices: Implications for Examinations, Findings, Evidence, and Decision Making

David A. Stoney, PhD, 14101-G Willard Road, Chantilly, VA 20151-2934*

After attending this presentation, attendees will appreciate the foundations for upcoming fundamental changes arising from the scientific examination of latent prints and the associated documentation, interpretations, and legal decision-making.

This presentation will impact the forensic science community by foreshadowing and accelerating fundamental changes attending the scientific examination of latent prints, increasing their utility, reliability, transparency, and fairness.

Gradually over many decades, latent print comparison practices developed and achieved a measure of standardization. The focus during this period was effecting identifications — a process whereby an expert in latent print examination and comparison reached a categorical conclusion that a latent print was, in fact, made by a specific individual. Measurement of latent print individuality, a prerequisite for scientific determination of significance, was not a part of this process. Efforts to measure individuality occurred over these many decades, but they were infrequent and were largely untested hypotheses. At no time were they part of the process used by practitioners to form their conclusions.

Crude, poorly defined counting of minutiae was frequently employed. These counts were sometimes used, following community practice or legal regulations, as a threshold for identification conclusions. Vagaries in counting criteria and the lack of scientific foundations led to international resolution for their abandonment in 1995.

As late as 1997, it was inconceivable to have a hearing challenging the admissibility of latent print evidence. In that year, the *West Companion to Scientific Evidence* was published, in which legal scholars solicited the collaboration of leading forensic scientists to articulate points of agreement and disagreement covering a wide range of scientific evidence. Other important developments were afoot, including improved biometric applications of fingerprints, application of these to the measurement and modeling of fingerprint individuality, legal scholarship challenging the historical foundations for latent print practices, and maturing evidentiary expectations for DNA-based identifications. Latent print identification practices, long ignored by scientists, legal practitioners, and the lay public, soon began to receive attention and started to become a focus for objective evaluation.

In 1999, it was no longer inconceivable to hold an evidentiary hearing on the admissibility of latent print evidence. This, and the resulting debate, directly motivated research into the related aspects of the measurement of latent print individuality. Over the next few years, methodologies used in latent print examinations were consistently challenged and consistently improved. Those who resisted the attendant changes most adamantly, or who held prior practices above reproach, were exposed by a combination of scientific scrutiny, legal awareness and high profile errors.

Over the past several years, important advances promoting these changes include:

- Continued legal scholarship and debate on the foundations of latent print examination, individualization conclusions, and errors.
- Research on the interpretation of physical evidence by applied statisticians and academic forensic scientists.
- Emergence of highly capable tools for performance and documentation of latent print examination protocols, applicable to: (1) initial latent print assessments of quality, reliability, and number of features for comparison; and, (2) comparisons of latent prints to candidate sources.
- Study of human factors in the latent print comparison process.
- Increasingly relevant and practical methods for the measurement of latent print individuality.

• During this last year or two particularly noteworthy are the publications of:

- A method for quantifying the weight of evidence from a forensic fingerprint comparison.
- Promulgation of professional guidelines and standards emphasizing transparency, documentation, and scientific practices.
- Explicit separation of the results of laboratory findings, from the evidence that results from their interpretation within case parameters and from their use in decision-making.
- Completion of an expert working group study of human factors

in latent print examination.

- A comprehensive, critical study of current methods used for the examination of latent prints, with recommendations for improving their reliability.

The impact of these events will be to re-focus issues in latent print analysis on the use of measurement tools in the latent print examination processes, together with their associated documentation. Measurement enables scientific progress. The result will be objective determinations of quality, sufficiency, and levels of complexity in the analysis. These, in turn, will dictate different required levels of documentation. The existence of explicit documentation will lead to transparency in the latent print examination process and the establishment of routine expectations for the examination itself and for informative discovery.

Latent Prints, Scientific Practices, Measurement

E6 Latent Print Testimony: What Lawyers Should Know to Ask and Examiners Should Know How to Answer

Heidi Eldridge, MS, LVMPD Forensic Laboratory, 5605 W Badura Avenue, Ste 120 B, Las Vegas, NV 89118-4705*

After attending this presentation, attendees will: (1) gain an understanding of some of the issues challenging fingerprint science in the courtroom in the wake of the 2009 National Academy of Sciences (NAS) Report, *Strengthening Forensic Science in the United States: A Path Forward*; (2) understand what some of the tough questions are that should be asked to ensure the expert is actually knowledgeable about his or her field and is using best practices; (3) understand how to recognize answers that represent outdated thinking and overstating of conclusions; and, (4) be able to recognize the expert who is thoughtful, transparent, and reliable.

This presentation will impact the forensic science community by providing lawyers with the inside knowledge they need to ask meaningful questions of the fingerprint expert and to recognize whether or not the expert has appropriate answers. Defense lawyers should be asking hard questions to ensure that any scientific evidence presented against their client is reliable, while prosecutors should be addressing these topics during pre-trial meetings to ensure that their expert is well-prepared and appropriately representing the science.

Since the 2009 release of the NAS Report, latent fingerprint examiners have been warned by various organizations, including the International Association for Identification (IAI) and the Scientific Working Group on Friction Ridge Analysis, Study, and Technology (SWGFAST), not to use certain phrases in testimony, particularly “zero error rate,” “to the exclusion of all others,” and “100% certainty;” however, many examiners are still testifying to these very things, while many don’t see any good reason why they shouldn’t. Part of the reason this is still happening is that lawyers are not asking the right questions. There is a pervasive feeling in the latent print community that “it can’t happen to me” — many examiners feel that they will never see these challenges and that there is no reason to change from “business as usual.”

Everyone needs to step up their game. Five years after the NAS Report, latent print examiners have had ample time to think about the criticisms of the report and find more appropriate ways to express their findings in court. Lawyers have also had ample time to find the questions that will force examiners to demonstrate an understanding of their own science and an ability to articulate it transparently and without exaggeration. Fingerprint science is reliable and it is probative. It does not require embellishment, but it does require thoughtful, transparent testimony that aids the trier of fact without misleading them through grandiose and unnecessary

claims.

This presentation will discuss advanced topics in latent fingerprint testimony. It will describe the reasons the above-mentioned phrases are not appropriate for use in fingerprint testimony and will demonstrate what more modest and transparent responses should look like. Concepts covered will include error rate, discriminability, specificity, uniqueness, individualization, and certainty. Attendees will learn about phrases that are commonly used, yet are not scientifically supportable and should not be permitted in expert testimony. Recent research that supports the reliability of fingerprint conclusions will be reviewed, along with its limitations.

Latent Prints, Testimony, Transparency

E7 “I No Longer Agree With My Trial Testimony:” The Legal Implications of Changed Expert Opinions

Michael Chamberlain, JD, CA Dept of Justice, 455 Golden Gate Avenue, Ste 11000, San Francisco, CA 94102*

After attending this presentation, attendees will gain understanding of the legal impacts and implications of an expert who, following trial, rethinks or otherwise comes to disagree with the opinion testimony that he or she gave in light of advancements in the field. Evolution of the theory and technology is, after all, synonymous with good science. The law; however, has a strong interest in finality and must resolve disputes and allegations fairly but quickly. How, then, does the law address experts who recant or modify their trial opinions after a criminal defendant has been convicted?

This presentation will impact the forensic science community by reassuring experts that courts do recognize the reality of evolving scientific theories and technology, but must also create workable standards to protect against wrongful convictions based on invalid scientific conclusions. It will describe how the California Supreme Court resolved these questions in a recent decision.¹ While *Richards* is a California case, its reasoning and holding should be of interest to forensic scientists nationwide, all of whom must face the issues considered in the case.

Richards involved the 1993 murder of Pamela Richards at the remote high desert home she shared with her husband William Richards. Pamela was strangled and her skull smashed with a cinder block. Forensic evidence included clothing fibers, DNA, blood patterns, footprints and tire tracks, and a bite mark. William was charged with her killing. Four trials ensued, with the fourth resulting in his conviction for first-degree murder and a 25-year-to-life prison sentence. Among the prosecution witnesses at the final trial was a forensic dentist who testified that a lesion on the victim’s hand was a human bite mark consistent with William as the biter. The expert further opined that William possessed unusual dentition because of a displaced tooth, and gave a population frequency estimate.

Ten years later, William Richards sought to have his conviction overturned with a writ of *habeas corpus*. He alleged that false evidence had been presented against him at trial, and that there was newly discovered evidence of actual innocence. Both claims were premised, in part, on a declaration by the dental expert that his trial testimony about statistical occurrence of William’s tooth pattern was not scientifically accurate, and that he could no longer say with certainty that the lesion on Pamela’s hand was a bite mark. He later stated at a hearing that William’s teeth were not consistent with the mark. Supporting declarations from other experts discussed advancements in digital photography since the

trial allowing for the correction of angular distortion in photographs of bitemarks and, consequently, more accurate comparisons.

The California Supreme Court decision will be discussed, including its acknowledgment that a changed expert opinion may be perfectly reasonable and need not imply a lack of integrity on the expert's part. Further, attendees will learn under what circumstances an expert's trial opinion can be shown to be objectively untrue, and when that finding will justify *habeas* relief. The example of SWGDAM DNA mixture interpretation guidelines—unrelated to the case—will be discussed. Finally, the fate of William Richards will be revealed in view of the court's reaction to his claims of false trial evidence and newly discovered evidence of innocence.

Reference:

1. Re: *Richards*, 55 Cal. 4th 948 (2012).

Expert Witness, Opinion, Recant

E8 Measurement Science and Standards in Forensic Handwriting Examination

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The goal of this presentation is to teach attendees about the current pertinent research relating to forensic handwriting examination and the future direction of research and case work. Attendees will also learn about the impact of this research on court cases involving forensic handwriting examination.

This presentation will impact the forensic science community by providing attorneys with solid case preparation materials in forensic document cases.

The Questioned Documents Section of the American Academy of Forensic Sciences (AAFS), the American Society of Questioned Document Examiners (ASQDE), the American Board of Forensic Document Examiners (ABFDE), the Federal Bureau of Investigation (FBI), and the Scientific Working Group on Forensic Document Examination (SWGDOC) all joined the Law Enforcement Standards Office (OLES) of the National Institute of Standards and Technology (NIST) as co-sponsors of the *Measurement Science and Standards in Forensic Handwriting Analysis Conference* on June 4-5, 2013, at the NIST campus in Gaithersburg, Maryland. All speakers were invited by the organizing committee to present on a tightly controlled theme. Quantitative measurements rather than qualitative aspects of forensic handwriting examination were emphasized. The program brought together forensic document examiners, researchers, statisticians, and an attorney to present the current state of the science and to design a roadmap for the future of forensic handwriting examination.

Forensic document examiners immerse themselves in significant texts and research in support of the science, but clients, attorneys, judges, and law enforcement personnel are uneducated about the complexity and scope of the work. This presentation will summarize the current research in support of forensic handwriting examination presented at this conference.

Since the *Daubert* decision was handed down by the Supreme Court, forensic handwriting examination has faced many judicial challenges, the most recent of which concerns the expression of similarity between the questioned and known specimens. Movement from a qualitative to a quantitative description of the expert's conclusions may give the courts more comfort.

Topics will include: the current state of handwriting examination; the foundational science behind handwriting identification, reproducibility and reliability studies; advances

in measurement science in handwriting analysis; advances in statistics for handwriting examination; the legal implications of quantitative testimony; and, other analysis tools to support metrics.

The future of the handwriting identification portion of forensic document examiners' work will include the expanding use and analysis of digital signatures. Research on electronically captured signatures by William Flynn and the use of acceleration/ deceleration plots in forensic analysis of electronically captured signatures by Kathleen Annunziata Nicolaidis will be discussed along with cases recognizing this work.

Forensic Document Examination, Measurement, Standards

E9 A Case Review of Conflicting Expert Opinions Based Upon Examinations of the Same Questioned Signatures and Exemplars

Andrew Sulner, MSFS, JD, Forensic Document Examinations, LLC, 220 E 57th Street, Ste 200, New York, NY 10022*

The goal of this presentation is to review a case in which the forensic examination and comparison of the very same questioned and disputed signatures led two groups of document examiners to reach conflicting opinions and to consider the factors that probably led to such disparate opinions.

This presentation will impact the forensic science community by showing how bias and motivational factors can lead experts to rationalize an outcome in the face of strong, if not irrefutable, evidence to the contrary.

Many disciplines in forensic science require experts to make subjective judgments about whether or not two visual patterns or sets of patterns are sufficiently similar to conclude that they originate from the same source. An extensive body of experimental research conducted by cognitive and social psychologists, as well as empirical data obtained from recent research and forensic casework studies, clearly demonstrates that various sources of bias can and often do adversely impact a forensic examiner's visual perception and decision making. As is the case in any pattern recognition task involving comparative analysis and evaluation by human observation and judgment, perceptual and cognitive judgments made by forensic document examiners performing comparative analyses of signatures or handwriting are susceptible to biasing influences that can improperly taint and sway the examiner's decision-making process, even without malicious intent.

Although the idea that bias can infect perception and decision making has been widely accepted by practitioners in virtually all scientific disciplines, forensic science practitioners involved in pattern recognition disciplines have traditionally dismissed bias as a significant factor influencing their perception and decision making, preferring instead to attribute conflicting expert opinions primarily to differences in the type of training received by the respective experts. The longstanding insistence on the part of most forensic examiners that practitioners with comparable training who *independently* examine the very same evidence will rarely, if ever, disagree with one another, is a myth that still prevails despite research, published decisions, case studies, and anecdotal evidence to the contrary. Far too many forensic examiners involved in pattern recognition disciplines such as fingerprints and handwriting still insist they are immune from being infected by bias because their fundamental education, training, and years of experience taught them to maintain total objectivity when making judgments about the evidence they examine and compare.

Conflicting Opinions, Questioned Signatures, Bias

E10 Where Eyewitness Identifications Go Wrong and Where We Go From Here: Case Study of Uriah Courtney

Alissa L. Bjerkhoel, JD, 225 Cedar Street, San Diego, CA 92101*

After attending this presentation, attendees will have a brief overview of the history of eyewitness testimony, its scientific validity, and cases where an eyewitness got it wrong. This presentation will use the case example of Uriah Courtney, whose conviction was based upon a bad identification.

This presentation will impact the forensic science community by serving to identify the very serious and continued implications of flawed eyewitness evidence as well as providing those in law enforcement and the legal community with guidelines to minimize misidentifications and prevent wrongful convictions.

Eyewitness identifications are the single leading cause of wrongful convictions.¹ There is almost nothing more convincing than a live human being who takes the stand, points a finger at the defendant, and says, "That's the one!"² Yet decades of research have proven that eyewitness identifications are often unreliable. Many times, the identifications are unreliable because of the methods traditionally employed by law enforcement when obtaining identifications. For example, studies show an increase in misidentifications where the investigating officer administers the identification procedure.^{3,4}

The exonerations of hundreds of American citizens prove, without a doubt, stranger eyewitness identifications are far from perfect. Of the nation's 311 documented DNA exonerations, 221 (or 71%) involved eyewitness misidentification.⁵ Roughly 35% of those exonerations involved misidentifications by more than one eyewitness.⁶ Between January and June of 2013 alone, DNA has proved the innocence of Garry Diamond, Joseph Frey, Jerry Jenkins, Robert Nelson, Bennie Starks, and Johnnie Williams. The most recent exoneration — Uriah Courtney — is a classic misidentification case.

November 24, 2004, would turn out to be no ordinary day for 16-year-old Erika. As she walked to her friend's house in broad daylight, a man came up from behind her, grabbed her, and told her not to scream. The man lifted her skirt and ripped her underwear. Erika fought vigorously with her attacker and broke free. The man caught her again, grabbed her, and threw her down in some bushes near a stoplight. There, he started to sexually assault her, but she eventually managed to escape into a passing car. She turned her clothing over to police and identified Uriah from a six-pick lineup as her attacker. The Sheriff's Department performed DNA testing on the victim's fingernail scrapings and her underwear, but did not obtain any meaningful results. Uriah was subsequently convicted and sentenced to life in prison.

The California Innocence Project started investigating Uriah's case and, due to recent understandings about the reliability of eyewitness identifications, garnished the cooperation of the San Diego District Attorney's Office. The victim's clothing was re-submitted for DNA testing and that testing revealed a male profile. That male profile was run through CODIS and a match was obtained to a local man who lived near the crime scene and had a striking resemblance to Uriah. Based on this new evidence, the parties agreed that Uriah should be freed.

In the United States, "[t]he vagaries of eyewitness identification are well known and the annals of criminal law are rife with instances of mistaken identification."⁷ The use of eyewitness identification evidence remains a crucial, yet troublesome, part of our criminal justice system. That being said, studies have shown ways in which errors can be minimized. Suggestions will be discussed.

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Eyewitness Identifications, Post-Conviction DNA Testing, Wrongful Conviction

E11 The Current State of the Admissibility of Bitemark and Other Pattern and Impression Evidence in Criminal Trials

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The goal of this presentation is to discuss the Innocent Project's (IP) highly publicized litigation around bitemark evidence over the past year and through that discussion offer insight on the admissibility of bitemark evidence and other pattern and impression forensic disciplines, now and in the coming years. More specifically, the presentation will discuss in detail a recent *Frye* challenge to the admissibility of bitemark evidence, focusing on the defense and prosecution litigation strategies and how this litigation will shape future challenges to the admissibility of all pattern and impression disciplines in the wake of the National Academy of Sciences (NAS) Report, *Strengthening Forensic Science in the United States: A Path Forward* and the FBI's unprecedented concession that its hair examiners have given scientifically invalid testimony in a still unknown number of cases over a period of decades.¹

This presentation will impact the forensic science community by elucidating current litigation strategies around pattern and impression evidence and the new limitations on the admissibility of such disciplines. A recent investigative report by the Associated Press, spurred by the IP's *Frye* challenge, documented 24 known wrongful convictions and/or arrests based at least in part on bitemark comparison evidence.² That report immediately preceded the FBI's concession that hair examiners had been overstating the probative value of an association between a known and a suspected hair for decades, leading to the re-examination of thousands of criminal cases.³ (Indeed, it was known prior to the concession that 72 innocent people have been convicted based on hair microscopy).⁴ The two hypotheses of both bitemark and hair comparison evidence — that: (1) a properly trained forensic analyst can make an association between a questioned sample and sample from a suspect; and, (2) a properly trained analyst can provide a scientifically valid estimate of the rareness or frequency of that association — have never been scientifically validated. The ever-increasing number of wrongful convictions based on bitemark, hair comparison, and other disciplines resting on essentially the same unvalidated hypotheses highlight the danger of admitting such evidence when life and liberty are at stake.

Over the past year, there has been a sea change in the

way courts and the forensic science community must consider the admissibility and probative value of bitemark and other pattern and impression forensic disciplines. The impetus for this change has its roots in the NAS Report but has accelerated in light of continued DNA exonerations, the reexamination of thousands of convictions based on hair comparison evidence and the IP's litigation against the admissibility of bitemark comparison evidence.

The court's decision in the *Frye* challenge, which will be rendered September 5, 2013, and the decisions that will emanate from the inevitable post-conviction litigation in hair comparison cases will be a referendum on the current state of the admissibility of bitemark comparison evidence, and, by analogy, other pattern and impression forensic disciplines. That is, will such evidence continue to be admitted at all? Or will its admissibility be significantly limited in how it is presented to juries? This presentation will be an opportunity to learn and discuss the future of bitemark evidence litigation specifically, and, more broadly, all pattern and impression evidence litigation in criminal trials.

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Forensic Odontology Bitemark, Hair Microscopy, *Frye/Daubert* Challenges

E12 What Is *Brady vs. Maryland* and Why Do I Care? A Review of Cases Discussing a Prosecutor's Duty to Disclose Exculpatory Evidence

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After attending this presentation, attendees will understand the facts and holdings of cases that impose a duty upon the prosecution to disclose to the defense information that exculpates an accused or that leads to impeachment evidence of important witnesses giving significant testimony. Attendees will also understand the significance of that duty for prosecutors in the context of consequence for appeals in criminal convictions.

This presentation will impact the forensic science community by discussing the prosecutor's duty to disclose exculpatory evidence. In the report by the American Bar Association (ABA) Criminal Justice Section's Ad Hoc Innocence Commission to Ensure the Integrity of the Criminal Justice Process, *Achieving Justice: Freeing the Innocent, Convicting the Guilty*, a recommendation regarding this body of law is made: "Prosecutors should ensure that law enforcement agencies, laboratories, and other experts understand their obligation to inform prosecutors about exculpatory or mitigating evidence." The intent of this presentation is to do just that.

During the commission of a robbery by two men, Brady and Boblit, a murder occurred. Felony murder, in Maryland, was

considered to be first-degree murder and punishable by death. The State of Maryland sought to convict each man of the capital murder offense and condemn each one to die. Of course, only one of the men actually pulled the trigger and he, Boblit, admitted to doing so before Brady's trial. Should his confession to killing the victim have been provided to Brady for his use, especially in the sentencing phase? The United States Supreme Court held that Maryland should have provided it to Brady and a new line of cases and codified rules regarding prosecutors' disclosure was begun.

Today, *Brady* and its progeny impose on the prosecution a "duty to learn of" as well as disclose to the defense all "favorable" material information known to the prosecutor and others acting on the government's behalf. This group is commonly referred to as "the prosecution team." The prosecution must disclose this information "at such a time and in such a manner as to allow the defense to use the favorable material effectively," because "the due process obligation under *Brady* is for the purpose of allowing defense counsel an opportunity to investigate the facts of the case and craft an appropriate defense."

Phrases that are frequently used demand a clear understanding for expert witnesses involved in an investigation and prosecution. What do those legal terms mean? What is "exculpatory?" What is "impeachment evidence" or "mitigating evidence?" Are there relevant factors to the prosecutor's duty to know about the information such as whether a witness is important or whether their testimony is significant? What is "significant?" Are personnel files of government experts information the prosecutor should know about? What about the type of an expert witness' findings in scientific testing of evidence in a case? What about "worksheets" that are created during the process of determining findings? How do existing cases answer those questions? What are those cases? Should knowing the answers aid experts in the application of them to unanswered questions? Should quality control information about a lab, competency information about the expert, or the findings of a peer reviewer be provided to the prosecutor?

Impeachment, Disclosure, Forensic Experts

E13 Error Reporting: Replacing Blame With Solutions

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After attending this presentation, attendees will understand the concepts of non-punitive error reporting systems and the possible obstacles to implementing such systems in forensic science facilities.

This presentation will impact the forensic science community by initiating a discussion of error reporting and the need to consider non-punitive error reporting systems to improve data collection.

"Error" — a five-letter word that provokes heated discussions in the forensic science community. Attempting to address "error" in the forensic sciences frequently leads to ideological arguments that divide our communities and result in accusations of information withholding, bias, and injustice. Even defining "error" can lead to accusations of bias. Narrow definitions of error, to support declarations that forensic science methods produce no "error," seem evasive and at times delusional. Broad definitions of error, to encompass all non-intentional or non-deliberate results, seem to condemn forensic science as faulty, incompetent, and unprofessional.

For all the accusations, heated arguments, and ideological divisions; however, we are a community united in our intentions —

we strive to perform good science and to get correct results. We want to remove “error” from our methods and our analyses. Yet, error occurs. Humans make mistakes. No system is perfect. Call them mistakes, errors, unintended consequences, or whatever euphemism is preferred, they occur. The relevant question is not what to call them, but how to avoid them.

In many professional fields, the trend has been towards non-fault or non-punitive error reporting to maximize data collection on errors. These fields include aviation, air traffic control, and various medical services. The stated goal in those fields has been to promote a “culture of safety” that seeks to increase reporting to prevent recurrences of errors and to generate information that could expose graver dangers (with more consequential effects). The systems rely on a commitment to shield reporters from any adverse or punitive consequences for both reporting the mistakes and making the mistakes. The systems apply to unintentional errors only; intentional errors can still lead to adverse consequences. Safety advocates have long touted the benefits of non-punitive systems to increase information that will lead to systematic improvements in procedures and the overall quality of services.

With the provision of forensic science services, maximizing the information about errors would seem to be a worthwhile endeavor. Learning from mistakes, improving operational procedures, and preventing scientific “disasters” are common, uncontroversial goals. But is implementing non-punitive reporting systems the right step for forensic science facilities and can a non-punitive reporting system function when forensic science services are provided for the adversarial legal system?

This presentation will discuss the features of non-punitive error reporting systems and whether the “culture of safety” dynamic supporting such systems in other fields can be transformed into a “culture of science” dynamic to support the systems in the forensic science community. The obstacles to using the non-punitive systems (which stress solutions) where results are introduced in legal proceedings (which are geared toward assigning blame) will also be discussed.

Error Reporting, Non-Punitive Systems, Culture of Science

E14 Forensic Laboratory Independence: “You Keep Using That Word — I Don’t Think It Means What You Think It Means”

Max M. Houck, PhD, Consolidated Forensic Laboratory, 401 E Street, SW, Washington, DC 20024*

After attending this presentation, attendees will have knowledge of the practical aspects facing forensic laboratories that are independent of law enforcement and the implications this has for relationships between scientists and attorneys.

This presentation will impact the forensic science community by providing real-world experiences of the first post-NAS independent forensic agency in negotiating the boundaries and shaping expectations for other independent laboratories.

Forensic service providers inhabit a necessary, if unique, role in the criminal justice system. The forensic sciences have many stakeholders, primarily the public they serve but also governmental ones, including law enforcement, attorneys, and the courts. For historical and political reasons, most forensic service providers are administratively a subset of law enforcement agencies. Occupying a subordinate role in a para-military organization sets boundaries on the laboratory’s relationships with their parent agency and aligned agencies. The “law enforcement” paradigm for forensic laboratories was challenged by the 2009 National Academy of Sciences Report (NAS) on the forensic sciences that recommended that forensic service providers be administratively or financially independent of

law enforcement-based parent agencies. The community response varied and concerns were raised: what about political clout during the budget process; who oversees the operations; how would the laboratory participate in investigations; and, what would be gained or lost through independence? Successful independence requires a greater sense of self-awareness and an objective analysis of the organization’s concept of operations. Without a framework or narrative in which to fit what a forensic science laboratory is (or isn’t), uncertainty will continue about the organization’s true mission, goals, and values. Relevant to the development of that concept is the notion of independence and what that means for a forensic laboratory. The question remains; however, how will the community move forward to evaluate and implement any new or adjusted business models for independent forensic laboratories? The need for strategic leadership in forensic science is critical and the lack of a historical systems-level view has slowed the development of strong strategic leadership. Forensic service providers, traditionally under law enforcement agencies, have had few opportunities to have a collective, distinctive political voice that served them alone.

As the first post-NAS independent forensic agency, the recent experiences of the DC Department of Forensic Sciences (DFS) may help illuminate some issues with independence, forensic service provision, and the role of forensic laboratories in the criminal justice system. The DFS interacts with the DC United States Attorney’s Office, the Public Defenders System, the DC Attorney General’s Office, and other law enforcement and legal agencies as an independent “science first” organization.

Independence, NAS Report, Laboratory

E15 Evaluating Quality in Crime Laboratory Casework: What Should Lawyers Know and How They Can Evaluate a Crime Lab

Barry A.J. Fisher, MS, MBA, 19854 Vintage Street, Chatsworth, CA 91311*

After attending this presentation, attendees will have a better understanding of ways attorneys will be able to evaluate forensic science lab reports and testimony proffered in criminal trials.

This presentation will impact the forensic science community by informing trial advocates of the ways forensic laboratories operate and explaining how to obtain important information through discovery and cross-examination to assist the court and jury in reaching a conclusion in criminal cases.

Consider a government forensic science laboratory that routinely provides testing services to the police and prosecution. Over time the laboratory’s clients implicitly trust the work product of the forensic lab. Challenges by the defense bar are expected but often not taken seriously. After all, in an adversarial criminal justice system, it is expected that the “other side” will try to diminish a crime laboratory’s findings.

In truth, there are scant ways for the police or prosecution to know if their forensic lab conducts reliable work. Laboratory clients have limited scientific expertise. They are, after all, police or lawyers and not scientists. How can they know if they are obtaining the sort of reliable, quality scientific evaluation the criminal justice system requires?

The burden to determine whether a forensic laboratory’s examination is accurate often falls on the defense. While police and prosecutors have a vested interest in the quality and reliability of a forensic lab’s work product, they may not know the proper questions to ask the expert or laboratory’s management if everything is okay or even if there are problems of which the prosecution should

be aware. In some cases, prosecutors and police investigators assume that all is well. And questions raised by lawyers on “the other side” are considered as unfounded issues.

Laboratory accreditation offers one way to evaluate the quality work product but it does not guarantee that the forensic science laboratory’s efforts are up to standard. Accreditation is an indicator. It’s fair to say that an accredited laboratory offers a better chance of quality analyses but accreditation cannot offer absolute guarantees.

Recently, we have seen examples of forensic labs that are part of police organizations that were not accredited. Not being accredited is not, *ipso facto*, proof of poor work, but it ought to raise questions to the parent law enforcement agency overseeing a crime lab: should our crime lab be accredited? Prosecutors might also wonder if something is not quite right. But what of the police agency or prosecutor’s office who doesn’t even recognize that the lack of accreditation or certification, or the existence of any quality assurance program, might be a harbinger of poor-quality forensic work? There are troubling instances where this, indeed, has been the case.

The prosecutor’s *Brady* obligation that requires them to provide exculpatory information to the defendant may help, but sometimes it will not. In some cases, labs and their personnel may not think to alert prosecutors about the crime lab’s shortcomings. Prosecutors may not recognize that a particular failing should be brought to the defendant’s attention or simply may not tell the defense. Thus, discovery becomes the vehicle for the defendant to evaluate a crime lab’s work.

The defendant is often at a disadvantage because he or she may not fully understand the day-to-day operation of a laboratory or simply not know the right questions to ask.

An American Bar Association (ABA), Criminal Justice Section taskforce recently drafted a resolution “*requiring laboratories to produce comprehensive and comprehensible laboratory and forensic science reports for use in criminal trials.*” The resolution made the following recommendations to include identification of: (1) the procedures used in the analysis; (2) the results of the analysis; (3) the identity, qualifications, and opinion of the analyst; (4) the identity and qualifications of those who participated in the testing including peer review or other confirmatory tests; and, (5) any additional information that could bear on the validity of the test results, interpretation or opinion.

These recommendations are a start. Defense attorneys should determine if labs use standard testing protocols which have been validated; if experts have been properly trained in testing procedures and take periodic proficiency tests; if lab reports are reviewed by qualified lab personnel before the case can be reported out; if labs maintain records of past errors and how the errors were corrected; etc. Understanding the accrediting process will help attorneys on both sides to determine whether forensic science used in a case is reliable and accurate and is helping, rather than hindering, the justice system.

Quality Assurance, *Brady* Material, Discovery

E16 Lessons Learned From Inside the Forensic Laboratory

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The goal of this presentation is to offer practicing lawyers insights about the inner workings of a forensic lab from the perspective of a former public defender, now General Counsel at the District of Columbia Department of Forensic Sciences.

This presentation will impact the forensic science community by bringing to the table the real experiences developing, growing, and changing as a forensic community as it strives to maintain independence as required by statute.

In October of 2012, the Department of Forensic Sciences opened in Washington, DC. The Department of Forensic Sciences houses forensics including DNA, firearms, fingerprints, and digital; public health; and crime scene sciences. By statute, the Department of Forensic Sciences is mandated to be an independent laboratory. Some parts of independence are included in the statute, such as the requirement of including two copies of discovery when discovery is requested, so that both sides have identical and equal access. A board of stakeholders, including the U.S. Attorney and the Director of Public Defense Services, is established. Likewise, a Scientific Advisory Board is established by statute, and approved by the District of Columbia’s City Council.

But other parts of “independence” haven’t been so easy. Who is “they?” Are police still “us?” What do we do when a prosecutor insists on testing items that have no forensic significance “so I can tell the jury we tested it?” How do we word reports? What value do we place on types of cases? How do we deal with contrary views of the value of evidence between police, prosecutors and the lab? When a defense expert wishes to observe testing, what level of vetting, if any, is appropriate?

The Department of Forensic Sciences took two unusual steps in establishing the Directorate at the lab. First, the Director decided that in-house counsel would better embody the Department of Forensic Sciences’ mandate for independence than a part-time attorney on call from the Attorney General’s Office. Second, the Director hired an attorney with 18 years’ experience — as a public defender. Benefits include a more realistic experience in moot court exercises, but also has sparked some interesting discussions such as the ones outlined above.

This presentation seeks to offer insights into how a having a lawyer in the crime lab can assist labs seeking to move toward independence. Additionally, it is hoped that this will open discussions between scientists and lawyers, seeking ways to improve the science — and the lawyering — that occurs in forensic science cases.

Independence, Forensics Laboratory, Lawyer

E17 Certification of Forensic Examiners in Texas

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After attending this presentation, attendees will understand: (1) the role of the Texas Forensic Science Commission (TFSC) with respect to the strategic advancement of forensic science; (2) the extent to which examiners in Texas were certified among participating laboratories; and, (3) efforts to address challenges associated with certification efforts.

This presentation will impact the forensic science community by raising awareness concerning the prevalence of certification among forensic examiners as well as the practical challenges and impediments to examiner certification that currently exist.

The mission of the TFSC is to strengthen the use of forensic science in criminal investigations and courts by developing

a process for reporting professional negligence or misconduct, investigating allegations of professional negligence or misconduct, promoting the development of professional standards and training, and recommending legislative improvements. In 2012, the TFSC sponsored an event entitled, "Strengthening Forensic Science in Texas: Moving Forward." A diverse group of forensic stakeholders was invited to discuss challenges and improvements that were broadly based upon the 2009 National Academy of Sciences Report, *Strengthening Forensic Science in the United States: A Path Forward*. As a follow-up to the Texas roundtables, the TFSC surveyed publicly funded accredited laboratories to determine the extent to which forensic examiners in Texas were certified, and to better understand the challenges related to certification efforts.

A total of 489 forensic examiners were represented in 22 publicly funded laboratories at the state (59%), county (23%), and city (18%) levels. Controlled substances (22%), forensic biology/DNA (17%), alcohol toxicology (15%), and firearms/tool marks (15%) were the most common disciplines or sections within the laboratory population, representing the largest numbers of examiners (167, 123, and 49, respectively). None of the publicly funded laboratories reported having sufficient examiners to maintain a 30-day turnaround in all disciplines in which they were accredited. The number of additional examiners needed to maintain a 30-day turnaround totaled 95 and ranged from 1 to 42 per organization. This represents a significant increase in scientific personnel of almost 20%. Of the 489 examiners, a total of 63 (13%) were certified. Two laboratories did not report certification by forensic discipline, but among the remaining 20 laboratories, the disciplines with the highest rates of certification were latent prints (21%) and firearms (16%). Certification rates among examiners in the most common disciplines, controlled substances and forensic biology/DNA, were 4% and 5%, respectively. The American Board of Criminalistics (ABC), the Association of Firearm and Tool Mark Examiners (AFTE), the International Association for Identification (IAI), and the American Board of Forensic Toxicologists (ABFT) were the most common certifying bodies. Support for certification among the forensic laboratory leadership was evidenced by the fact that 50% of the participating organizations already offered some form of incentive for examiners to become certified. Most of the laboratories support certification or view it as inevitable, but also recognize the formidable challenges associated with this effort in terms of funding, training resources, and personnel.

Forensic, Certification, Commission

E18 Improving Fire Investigations Through Partnerships, Case Reviews, and Training

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After attending this presentation, attendees will have learned how to improve fire investigations by: (1) developing partnerships with various stakeholders; (2) ensuring justice through forensic reviews; and, (3) developing a strategic approach to training.

This presentation will impact the forensic science community by describing the efforts of the Texas State Fire Marshal's Office (SFMO) to put into action the recommendations found in the 2011 Texas Forensic Science Commission (FSC) report on fire investigations. The fire-investigation challenges described in the FSC report are not unique to Texas, and the judicial system must demand that these investigations be conducted in accordance with national standards, rather than relying on myths passed from generation to generation of fire investigators. The

term "junk science" is often used with reference to theories that have been — and continue to be — utilized in fire investigations, but in fact lack any scientific basis. Attendees will learn how to implement a systematic approach to improving fire investigations in their organization and community.

The Texas SFMO established a unique partnership with the Innocence Project of Texas (IPOT) and the FSC to make sure that fire investigations are based on science and best practices. There was a recognized need to restore public confidence in the quality of fire investigations, and the approach of the Texas SFMO has been to be transparent in its partnership with IPOT and the FSC as changes are implemented. This partnership has worked very well, since the only agenda is to make sure that justice is served by conducting fire investigations based on solid scientific methodologies.

The involvement of science experts in fire investigations, especially those conducted by public-sector investigators, is a significant issue that needs to be addressed. The participation of science experts in public-sector investigations has traditionally been very limited. The research into how public-sector fire investigators incorporate the scientific community into fire investigations did not yield any programs to model; this required creation of a forum for training, consultation, and case reviews. The creation of the Texas SFMO Science Advisory Workgroup has produced a panel of experts in the fields of forensic science, chemistry, electrical engineering, mechanical engineering, forensic pathology, law, and fire investigations. In Texas, these experts provide quarterly training to fire investigators, perform retroactive reviews of cases (including those presented by the IPOT), and serve as consultants to the Texas SFMO. This workgroup began work in January 2013 and issued its first findings on three cases in June. In one case, the panel supported the original fire investigation, but it found deficiencies in the other two investigations, concluding that the respective fire causes should have been ruled as undetermined. The local District Attorney and Texas Court of Criminal Appeals have received these reviews and will decide what needs to be done with regard to the two cases that did not meet current fire investigation best-practice standards. It is important to note that the Science Advisory Workgroup only looks at the science of a fire investigation and does not examine any peripheral issues, such as the materiality of the fire investigator's conclusions to the final outcome of a particular criminal conviction. The Workgroup leaves those determinations to the appropriate parties within the criminal justice system.

Training is critical in order to meet best-practice standards, and there must be a strategic plan to meet NFPA 921 and 1033 standards, and to ultimately exceed these minimum requirements. In the public sector, this is a significant challenge that requires coordination among various stakeholder groups. Partnerships are the key to providing excellent, cost-efficient training.

The status quo in the field of fire investigations must change in order to dispel public perceptions of investigators' use of "junk science" and to align the profession with proven forensic science.

Arson, Review, Partnerships

E19 Improving Forensic Science Through State Oversight: The Texas Model

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After attending this presentation, attendees will understand how state forensic oversight bodies such as the

Texas Forensic Science Commission (“Commission”) can play a critical role in improving the integrity and reliability of forensic science in a state’s criminal justice system. The history and legal authority of the Commission and the critical nature of its make-up (seven scientists, one prosecutor, and one defense attorney) will be presented. Attendees will also understand the Commission’s approach to investigating complaints and self-disclosures. It will be demonstrated how the Commission’s ability to be flexible and adapt quickly to developments in the forensic community are key to successful oversight, especially when contrasted with the relatively slow pace of reform at the national level.

This presentation will impact the forensic science community by demonstrating the tremendous potential for state leadership in forensic science oversight and by encouraging attorneys, forensic scientists, policymakers, and other attendees to be creative and proactive in tailoring state-based forensic science oversight programs. This presentation will provide concrete examples of how states can move reform initiatives forward in a flexible and collaborative manner and at a robust pace.

The importance of a strong crime laboratory self-disclosure program will be demonstrated, including ways to encourage laboratories to come forward with facts that may form the basis of negligence or misconduct in a crime laboratory. The presentation will discuss how shared statewide expectations regarding the role of a forensic science commission can increase transparency and improve organizational culture. This presentation will emphasize how much progress can be made when a forensic oversight commission insists upon comprehensive yet fair investigations, resists external political agendas, and adopts a collaborative approach to reform with all affected stakeholders.

The presentation will also discuss the importance of proactive interaction between a state commission and the national accrediting bodies, using ASCLD-LAB as an example. There will be an emphasis on the critical nature of distinguishing between the role of an accrediting body and the role of a commission that conducts investigations into negligence and misconduct. By using examples of cases investigated by the Commission in various forensic disciplines (e.g., controlled substance analysis, forensic biology, etc.) as well as institutional broad-based reviews (e.g., arson investigation, hair comparison, etc.) attendees will better understand how state commissions can facilitate meaningful forensic review when scientific developments and ethical obligations require it.

This presentation will also focus on the importance of a state commission’s outreach to affected prosecutors, local government officials, and the defense bar *during the course of a given investigation* to ensure the lawyers and responsible government officials understand the implications of a particular forensic nonconformance. Attendees will better understand the Commission’s efforts to develop a statewide notice protocol to ensure potentially affected defendants receive notice in the wake of a major forensic failure involving a high-volume discipline such as controlled substance analysis.

Finally, by highlighting the Commission’s collaborative training efforts with the Texas Criminal Justice Integrity Unit, it will be demonstrated how targeted training initiatives can improve the long-term understanding and integrity of forensic science throughout the criminal justice community.

Forensic, Commission, Model

E20 Plausible Deniability — The Ethics of Inconsistent Consistency

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After attending this presentation, attendees will have a better understanding of the nature of qualifiers and vague terminology in the process of overstating and understating opinions in court.

This presentation will impact the forensic science community by bringing to light common vague and/or unclear terms to provide a better understanding of the nature of such qualifiers and vague terminology in the process of overstating and understating opinions in court.

One of the singularly most critical components in science is often one of the most overlooked — effective communication. A fount of wisdom slakes no thirst if none can drink the waters of knowledge. The tipping point in forensic science, the 2009 National Academy of Sciences Report, specifically addressed the issue of understandable terminology

Certain terms are specifically criticized as being unclear means of conveying useful information. For example, the term “match” appears 38 times, usually associated with pattern evidence and specifically fingerprint identification. Yet in the entire report, the forensic catch phrase “consistent with” is mentioned in the same light only once: *The meaning of “exceeds the best agreement” and “consistent with” are not specified, and the examiner is expected to draw on his or her own experience.*¹

In that same NAS report, the term “consistent with” is specifically cited in the context of improving the science of forensic science — three times — without any mention of confusion or need for clarification. This apparent dichotomy of critique and application begs the questions of how truly effective is information exchange between the various parties and if a possible double standard exists. The result can be that in the highly partisan world of the justice system, witnesses sometimes become involved in the tautologies of law, when, in fact, the critic may be equally, if not more, culpable. Akin to this issue are the terms “reasonable certainty” and “reasonable probability” which are routinely encountered in both civil and criminal justice fora. Equally frustrating can be encounters with experts who intentionally or otherwise obfuscate the truth with qualifiers to the extent that one is left uncertain of specifics about an opinion in a case under discussion. This can ultimately lead to high-profile “battles of the experts” which may make for increased media ratings but does precious little to advance science, understanding, or the quest for justice.

Reasonable and ethical scientists can agree to disagree — no human holds the ultimate authority of absolute truth. Instead, cases often revolve around perspective issues — how findings are interpreted and challenges are presented. Condemnations may be made not only of specific observations but also of omissions. One party may criticize another’s “obvious” failure to give proper consideration of certain “key elements” of a case, in an attempt to bolster acceptance of an alternate opinion. The dangers lie in the fusion of observation and opinion. While still seeking a fundamental higher ground of seeking facts, the aspirant does so based in part on training and experience. Specialists in a particular endeavor might carry a subliminal bias based in part on the observational nature of the task — one “finds” what one “seeks.” Ultimately, disputes about cases may come down to opinions regarding factual observations and how those same assessments are received in court.

A multidisciplinary panel will utilize a case-study approach,

discussing several cases where different expert opinions were in sharp contrast, yet ultimately only one was accepted by the court. A common theme encountered is overstating the general strength of opinions and the ability to reach certain conclusions. Fundamentally, a broad qualifier such as "consistent with" attempts to cover a broad swath of foundational information. The ethics of over-reliance of such vagueness at the expense of a systematic analysis will be debated, with examples of the eventual adverse impact of such techniques — both on individual cases and in the broader context of professionalism in forensic sciences.

Reference:

1. <https://www.ncjrs.gov/pdffiles1/nij/grants/228091.pdf>

Testimony, Consistent With, Ethics

E21 Homicidal Violence: Is It an Admissible Cause of Death?

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After attending this presentation, attendees will be familiar with the criteria for the certification of deaths due to homicidal violence and the legal concerns regarding the designation.

This presentation will impact the forensic science community by the recognition and means of certifying the manner of death in difficult cases. It will also prepare judges and other legal practitioners to understand the criteria for the certification.

In 2008, Donald Cooper, the leader of a Milwaukee drug gang was convicted of two counts of murder in the deaths of two victims found buried under concrete slabs. The jury convicted Cooper despite the fact that the medical examiner was unable to definitively determine a cause of death and instead had certified the death as due to "homicidal violence." In Detroit in 2013, James Brown was convicted of the deaths of four women found locked in automobile trunks who were certified as homicides with no identifiable injuries.

The primary role of the medical examiner or forensic pathologist is to determine and certify the cause and manner of death. On occasion, the exact cause of death cannot be determined within a reasonable medical probability despite the presence of overtly suspicious circumstances and evidence associated with the death. In these cases, medical examiners are likely to certify the death as homicide by unspecified means or simply homicidal violence. In a recent article by Matshes and Lew, the authors developed the following criteria for the certification of a homicide by unspecified means.¹ The criteria include cases, in which, following a complete autopsy, there are: objectively suspicious circumstances of death; no anatomic cause of death; no toxicological cause of death; no historical, environmental, or circumstantial cause of death; and, when a more specific cause of death cannot be suggested by the dataset. The criteria specifically referred to cases where the body had deteriorated, destroying vital evidence and circumstances directly relating to the death.

Although the certification of homicide by unspecified means is accepted by most vital records departments, it remains controversial in the courtroom. Forensic pathologists adhere to a standard of proof as a "reasonable degree of medical probability," well below the "beyond a reasonable doubt" standard of the jury. The lack of a definitive cause of death may severely impact the jury. The determination of a death solely on circumstances, it could be argued, invades the purview of the jury and should therefore not be accepted as an expert opinion by the court. Others argue that the determination of the cause of death, even when the cause cannot be definitively demonstrated, rests on expert's education and experience.

This presentation provides case presentations certified as homicidal violence resulting in judicial convictions. A discussion of the definition of homicide by unspecified means will be presented as well as concerns of judicial acceptance and the role of the medical expert.

Reference:

1. Matshes E. and Lew E., "Homicide by Unspecified Means," *Am J Forensic Med Path*, 31 (2), 2010:174-178.

Homicidal Violence, Death Certification, Evidence Admissibility

E22 Shot in the Back: A Case Study of a Murder Investigation in Indiana

Christine Haskell, JD, 128 N Cullen Street, Rensselaer, IN 47978*

After attending this presentation, attendees will understand the importance of law enforcement officers and prosecutors working together during an investigation through the collection of evidence at the crime scene, the processing of the forensic evidence collected, the release of information to the public regarding the investigation, and the prosecution of the accused.

This presentation will impact the forensic science community by providing a case example of the importance of collecting forensic evidence by law enforcement officers and coordinating with the prosecutor on the progress of the investigation. In addition, it will demonstrate how the release of certain information about an investigation prior to the analysis of evidence obtained can interfere with the prosecution of the accused. The case example will illustrate how vital it is for the law enforcement officers, forensic experts, and prosecutors to collaborate from the first moment a murder is reported.

The primary goal of this presentation is to present a case study of a homicide investigation in a rural county in Indiana. Jasper County, a rural northwest county in Indiana with a population of less than 40,000 people, has had less than ten reported homicides over the last 25 years. In Jasper County, law enforcement officers and prosecutors rarely have the opportunity to investigate homicides. Therefore, when a homicide investigation arises, the investigation has the potential of becoming problematic based on the lack of frequency with which they occur.

In this particular case study, law enforcement officers responded to a shooting at the home of a man and his son. When they arrived, the father was found lying dead on the front porch, with a gunshot wound to his back. The son told the officers that he shot his father in self defense and in defense of his girlfriend, whom his father had been sexually assaulting. A crime scene technician arrived and processed the scene and collected forensic evidence for analysis. Law enforcement officers continued to investigate the crime by questioning witnesses about what they observed. Based on the initial information, the law enforcement agency issued a press release regarding the alleged crime and the suspect, before all elements of the case were in. This press release was issued without consulting the prosecutor's office and prior to the crime scene technician providing his report. Of significance in the press release was the statement that the shooting was the result of a domestic dispute that started because of the father's sexual advances on the girlfriend. Eventually, law enforcement officers and the prosecutor assembled to discuss the investigation and ensure that necessary evidence was obtained that had previously not been considered, such as DNA analysis of the girlfriend's clothing, toxicological results of both the suspect and the victim, and retrieving phone records of the suspect. When the evidence and statements were obtained, they presented evidence that contradicted the story of the accused.

Law enforcement and prosecutors learned many valuable lessons on how to investigate a crime, the necessity of being informed of the investigation, and the importance of when to release information about the investigation. In response to the case, a policy was established for responding to a murder investigation and how the collaboration of law enforcement, prosecutors, and forensic experts is essential.

Criminal Justice, Evidence Collection, Case Study

E23 Cutting-Edge Legal Issues and Considerations Surfacing From the Scientific Process Involving Hydraulic Fracturing (Fracking)

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After attending this presentation, attendees will gain an understanding about the legal issues, controversies, and considerations involving fracking.

This presentation will impact the forensic science community by providing statutory and case law evaluations on the legal issues regarding fracking. The case law in this area is still developing, and the legal issues are cutting-edge issues demonstrating the tension between the interests of the parties — the landowners, the environment, and the natural gas industry.

One of the biggest shale deposits in the United States, the Marcellus Shale, which is located under much of Ohio, West Virginia, Pennsylvania, New York, and small regions of Maryland, Kentucky, and Tennessee, is worth approximately \$500 billion. Fracking is the process of extracting natural gas from gas shale deposits located approximately a mile below the surface and this process has resulted in massive new supplies of oil and clean-burning natural gas that have increased our country's energy security and have improved our ability to generate electricity, heat homes, and power our vehicles for generations to come. Fracking has contributed to boosting many local economies through royalty payments to landowners, tax revenues, and job creation. As a result of this massive production potential, many legal issues, controversies, and concerns are surfacing around how environmental laws are created and how they should be enforced. There are heated jurisdictional controversies regarding the application of federal versus state jurisdiction and whether the state or federal government should have the final word as to the outcomes pertaining to specific environmental issues related to the oil and gas industry. An overlap of laws in this area is created by the federal government, the state government, and the municipalities at a local level of government.

Several states such as Colorado, Texas, Pennsylvania, Louisiana, and Wyoming allow and regulate fracking in each of their own ways. Other states like New York, as of this writing, have not approved fracking, and yet some New York state courts have taken preemptive measures to limit or prevent fracking. The perspectives of each of these states and other states will be examined.

Currently, the most common litigation involves title issues surrounding the ownership rights of minerals extracted from real property. Many of the more cutting-edge legal issues are related to the Halliburton Loophole, in that there have been efforts to introduce legislation to remove the oil and gas industry exemptions from the federal Clean Air Act and the Clean Water Act. Such legislation specifically seeks to remove exemptions related to storm water run-off at drill sites and emissions generated by the oil and gas

industry.

A recurring issue is determining whether fracking activities contribute to or are responsible for contaminating drinking water; however, studies appear to be confirming that fracking happens so far from groundwater, with so much rock in between, that it is unlikely that fracking is resulting in the pollution of groundwater. Of more concern are the issues surrounding waste water disposal. Currently, Pennsylvania is pushing drillers to recycle waste water, a process which is also stimulating the economy and creating new jobs. In addition, structural concerns exist regarding older wells being retrofitted to comply with new regulations.

Another legal issue in this area is the sealing or unsealing of court records regarding details of settlement. After landowners sue the natural gas companies for damages for alleged contaminated waters, the parties, especially the corporations involved, are requesting the trial courts seal the terms of the settlement so that the public is not aware of the precise amount of settlement. The media outlets are filing petitions to intervene to unseal the court records. In this age of transparency of government records and the constitutional presumption of openness in judicial proceedings, the right of privacy or "secrecy" involving the settlement amount must be analyzed and weighed against the public's right to know the precise settlement terms. The timeliness of the intervention is another legal concern.

In conclusion, this presentation will provide the legal issues and concerns regarding hydraulic fracturing which has become a major jurisdictional issue and involves other developing issues attracting a significant amount of attention in the near future for, with, and by our judiciary.

Fracking, Legal Issues, Scientific Process

E24 The History of DNA Evidence and the Rule of Law: Science and the Law Three Decades Later — Will the Law Ever Catch Up?

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The goal of this presentation is to carry out a historical analysis of the development and utilization of forensic DNA testing as it occurred in the United States and to demonstrate the causal interplay and corresponding change to significant legal doctrines in the American judicial system.

This presentation will impact the forensic science community by providing an overview of the history of DNA evidence and the cause-and-effect relationship with changes in our law. An understanding of the history of DNA will give attendees insight into coming shifts in legal principles that are sure to ripen in the future.

The use of DNA evidence has had a profound effect on the adjudication of cases within our adversarial system of justice. As a product of the unique power of DNA testing to correctly resolve factual issues, long-held legal principals have been re-examined, both legislatively and through the decisional law. Statutes of limitations, scientific admissibility, and the doctrine of finality of cases are a few examples of areas of law that have been affected by DNA science being introduced into the courtroom.

Forensic science is simply defined as the application of science to the law or legal matters. When the judicial system needs science to resolve a question, the person who is called upon to bring science into the courtroom is a "forensic scientist." Science is an empirical method of learning, anchored to the principals of observation and discovery as to how the natural world works. Scientific knowledge increases human understanding by

developing experiments that provide the scientist with an objective answer to the question presented. Through the scientific method of study, a scientist systematically observes physical evidence and methodically records the data that supports the scientific process. The law, on the other hand, starts out with at least two competing parties who use the courthouse as a battleground to resolve factual issues within the context of constitutional, statutory, and decisional law.

DNA analysis has set a high standard against which other forensic sciences are now being judged. Not only has DNA identity testing redefined the standard of acceptability of other scientific evidence, it has also fostered an awareness among juries that non-DNA-based identification techniques are less supported scientifically and therefore should be less accepted than DNA profiling as a method of scientific investigation. The 2009 National Academy of Sciences Report was critical in its assessment of some forensic disciplines.¹ Lack of research supporting the basic tenets of techniques was noted. The gist of the NAS Report was that the admittance of a scientific technique into the courtroom when there is very little to support its validity can have consequences that are potentially disastrous. A number of exoneration cases have exemplified the errors that can be made. When life and liberty are at stake, there is a responsibility to base scientific testimony on substantiated techniques. When testimony is opinion based and not science based, interpretation can become subjective rather than objective.

A working knowledge plus an understanding of the advance of forensic DNA identification science is important to all forensic scientists and attorneys who practice in any field of the forensic sciences. The catalyst for DNA's effect on the American legal system was the development and acceptance of DNA identification genetic testing which began in the 1980s. The use of DNA took firm root in the 1990s and was entrenched by the early 2000s. DNA is considered to be the proverbial "gold standard" of biological human identification. DNA profiling over the past three decades was the most significant advance in forensic science since the development of fingerprinting in the 1900s. New types of DNA are being evaluated along with related technologies, notably the continued development and expanded use of DNA data bases. These new developments will continue to make DNA identification an ever more momentous stimulus to change the legal system. Soon "Rapid DNA testing" technology will be emerging. The DNA revolution was a sea change in how courts comprehend what is science. New DNA developments will continue to have vast implications to the rule of law.

Reference:

1. A published study of the National Research Council entitled *Strengthening Forensic Science in the United States: A Path Forward*. The National Academies Press, 2009, hereafter NAS Report.

DNA, RAPID DNA, History

E25 DNA and Voir Dire: What Does Your Jury Already Know About DNA?

Julie Maxwell, JD, 510 15th Street, NW, Rochester, MN 55901*

After attending this presentation, attendees will gain insight into how to approach questioning prospective jurors regarding the DNA evidence in their cases. In the jury pool, there will be not a single, common concept of DNA. Some of the prospective jurors have no idea what DNA is, much less what impact DNA evidence can have on a criminal case. For other prospective jurors, their only knowledge of DNA is what they have seen on TV crime shows. Some may have the belief that the presence of a defendant's DNA

is absolute, complete, and total proof of his/her guilt. A few may have a little more realistic grasp of what DNA evidence is, how it is obtained, and how it can be useful in a criminal case. *Voir dire* is the best way to help the attorney determine just how much knowledge the prospective jurors bring to courtroom. Examples of several different types of *voir dire* questioning will be presented.

This presentation will impact the forensic science community by showing how much impact carefully crafted *voir dire* questions can have in a trial. Attendees will see what lines of questioning can be most productive in gathering information about the prospective jurors' beliefs, what lines of questioning do not provide as much useful information, and what lines of questioning to try to avoid because they could end up being counterproductive and confusing to the prospective jurors.

The NAS Report found that lawyers "often lack the scientific expertise necessary to comprehend and evaluate forensic evidence." In response, the Minnesota State Public Defender, working with a small group of attorneys, developed a year-long training program to teach 30 assistant public defenders DNA-typing litigation skills, using a combination of intensive lecture, small group discussion, and one-on-one tutoring. Each attorney applied this advanced training to one of their own actual, pending cases. Prior to this training, many attorneys just accepted the reports submitted by the Minnesota Bureau of Criminal Apprehension for their face value. Now, following the training, nothing is taken at face value. These lawyers have increased confidence in their ability to spot issues and then to work with experts on complex forensic issues.

To further sharpen the trial skills of this group, the Minnesota State Public Defender offered an opportunity for three of these DNA Institute attorneys to attend its annual one-week Trial Advocacy School. Each of the DNA attorneys worked with a non-DNA attorney on a case in which DNA played a major role. *Voir dire* was among the trial skills which were the focus of this training opportunity. Volunteer prospective jurors assisted in this exercise. Each DNA attorney prepared questions specifically designed to learn how much DNA information the prospective jurors had prior to the exercise. The attorneys also received feedback from those jurors concerning the effectiveness of their questions as well as feedback from instructors. The attorneys then had the opportunity to modify their questions and repeat the exercise, if they chose to do so.

This presentation will give attendees examples of how this additional advanced trial training for three members of the Minnesota Public Defender Advanced DNA Institute can lead to improved performance during jury trials.

Voir Dire, DNA, Questioning

E26 DNA Mixture Interpretation: History, Challenges, Statistical Approaches, and Solutions

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After attending this presentation, attendees will better understand the DNA mixture interpretation approaches used in forensic DNA laboratories today.

This presentation will impact the forensic science community by helping the legal community appreciate laboratory difficulties with DNA mixture interpretation.

Since its introduction in the mid-1980s, forensic DNA testing has played an important role in the criminal justice community through aiding conviction of the guilty and exoneration

of the innocent. New technologies are regularly introduced and validated to expand the capabilities of laboratories working to recover DNA results with improved sensitivity and informativeness. One of the largest challenges today is coping with interpretation of complex mixtures and low-level DNA profiles where portions of the evidentiary profile may be missing and thus unavailable for comparison to reference profiles.

A brief history of the forensic DNA field will be provided with a review of approaches to DNA mixture interpretation. Due to the prevalence of mixtures in many forensic casework situations, multi-allelic Short Tandem Repeat (STR) markers will likely remain a primary workhorse for DNA analysis into the foreseeable future. The 2010 Autosomal STR Interpretation Guidelines from the Scientific Working Group on DNA Analysis Methods (SWGDM) have led to protocol changes in many forensic DNA laboratories.¹ The role and limitations of stochastic thresholds that are commonly used with some statistical methods will be discussed. The 2010 SWGDM guidelines were written with a focus on single-source and two-person mixtures, and limitations exist in applying some basic concepts to more complex mixtures. Lessons learned from NIST interlaboratory studies will be reviewed along with information available on the NIST STRBase website that relates to mixture interpretation (<http://www.cstl.nist.gov/strbase/mixture.htm>). Key literature references and other educational resources will also be discussed.

Several software programs enable statistical calculations to be performed with probabilistic genotyping and/or incorporating a probability of allele dropout. These approaches enable analysts to account for the possibility of missing data in complex or low-level evidentiary DNA profiles. In December 2012, the DNA Commission of the International Society of Forensic Genetics (ISFG) published recommendations on evaluation of STR typing results that include drop-out and/or drop-in using probabilistic methods.² Some examples will be shared to show the relevance of different approaches that can be taken when complex DNA profiles are present in evidentiary results.

References:

1. http://www.swgdam.org/Interpretation_Guidelines_January_2010.pdf
2. Gill, P., et al. (2012). DNA Commission of the International Society of Forensic Genetics: Recommendations on the evaluation of STR typing results that may include drop-out and/or drop-in using probabilistic methods. *Forensic Science International: Genetics*, 6, 679-688. Available at <http://www.isfg.org/Publication;Gill2012>.

Forensic DNA, DNA Mixtures, Mixture Interpretation

E27 It's Rapid, But Is It Relevant? Balancing Speed and Evidentiary Significance in the Coming Age of Real-Time DNA Analysis

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After attending this presentation, attendees will understand why test results obtained from rapid DNA technologies must not significantly outpace investigative information about the items from which those results are obtained. It is critical that police agencies quickly discover, document, and disseminate information about such items to establish their relevance as evidence. Only then can rapid testing technologies truly benefit law enforcement in the coming age of real-time DNA analysis.

This presentation will impact the forensic science community by explaining why rapid DNA results will be but a single

component of a tested item's evidentiary significance. It will also explain why such results are meaningless absent investigative information that builds a contextual framework around a rapidly produced profile. This factual context, in turn, animates evidence with varying degrees of logical relevance. Accordingly, the rapid DNA revolution will never be fully realized without concurrently improving the timely collection and transmission of item-specific information to essential investigative assets. Only then will the right evidence be tested, providing real-time results with real-time relevance.

Physical evidence is composed of three separate elements: (1) the relative degree of significance inferred from an item's contextual surroundings at a crime scene; (2) what those in a position to know — witness(es), victim(s), or suspect(s) — say (or do not say) *about it*; and, (3) the forensic testing results, conclusions, and weight (quantitative or qualitative) of a match or association to that item.

These three elements correlate with the work performed by three types of investigative assets employed by most police agencies — crime scene investigators (element 1); detectives (element 2); and, forensic scientists (element 3).

Differences in expertise, specialization, and division of labor necessarily require these investigative assets to simultaneously work on different aspects of the same case, gathering information about crime scene evidence from multiple sources and locations. As a result, information about physical evidence is not routinely shared — in real-time — among these assets as it is acquired. Accordingly, crime scene investigators may collect numerous items for DNA analysis before a factual nexus between those items and the case has been established. Alternatively, scene investigators may fail to collect items that ostensibly appear to be insignificant. In reality; however, they may have a direct association with critical, but then, unknown, case facts.

Detectives are normally responsible for interviewing victims, witnesses, and criminal suspects. Typically; however, these individuals are promptly removed from the scene and taken to a separate location to be treated or interviewed. Consequently, interviewing detectives may never enter the crime scene and thus remain unaware of the nature and significance of the physical evidence it contains. Furthermore, in an effort to quickly develop a lead and make an arrest, detectives are likely to focus on questions concerning “whodunnit” rather than “howdunnit.” As a result, they may fail to acquire critical information about the relative significance of specific crime scene items from persons with such knowledge.

Forensic scientists receive evidence analysis requests from detectives who may seek DNA testing of one or more (and possibly dozens of) submitted items. In complex investigations; however, detectives may have little, if any, information about *why* crime scene investigators collected certain items or samples, *how* they relate to the case, and *when* they were deposited at the scene. Furthermore, in the haste to develop a rapid genetic profile, DNA analysts may bypass preliminary serological testing that would further deprive a detected profile of contextual significance.

As a result of these investigative disconnects, the relevance of rapidly produced profiles may be uncertain in many cases. Therefore, coordination and communication regarding physical evidence must be greatly enhanced to meet the coming age of real-time DNA analysis. This can be accomplished in a number of ways.

First, crime scene investigators, while still at the scene, must communicate with detectives and share specific information about the presence and nature of potential items of evidence. This may generate further investigative questioning by detectives. Additional questioning may, in turn, lead to the identification, collection, and/or processing of additional relevant items by scene investigators.

Second, in addition to questions focused on suspect development, detectives must begin to routinely ask victims and witnesses about which particular items or samples at a scene may be significant to the investigation, and why that is the case.

Third, relevant interview information, whenever possible, should be relayed by detectives, in real-time, to crime scene investigators. This will allow them to identify, collect, and/or process items that may otherwise appear to be insignificant or unremarkable.

Fourth, crime scenes should *not* be released until *after* detectives have acquired all necessary information from victims, witnesses, and suspects about potentially relevant items of physical evidence.

Fifth, item-specific information must be collated, documented, and disseminated by investigators to DNA analysts who can determine if serological testing is advisable, given the nature and context of the sample. Furthermore, when more than one item is submitted, analysts, case detectives, and prosecutors must collectively make reasoned judgments about testing prioritization based on each sample's potential for successful DNA analysis and its relative degree of probative value.

In summary, the results obtained from rapid DNA testing are but a single element in the tripartite analysis of evidentiary relevance. Test results that outpace law enforcement's acquisition and dissemination of basic information about questioned crime scene items are not fully useful. In such cases, law enforcement merely has a profile waiting on a personality. Therefore, in addition to profile speed, it is essential to achieve *rapid relevance* for real-time results.

Rapid DNA, Relevance, Evidence

E28 “Detective, I Swear I’ve Never Seen That Woman Before:” How a Killer’s Denials Plus DNA Put Him Away for Life

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After attending this presentation, attendees will learn how new DNA collection methods enabled criminalists to develop evidence from fingernail clippings collected in a 25-year-old unsolved homicide.

This presentation will impact the forensic science community by illustrating how a new method employed by the OCME of NYC developed evidence that led to the reopening of a cold homicide case, and how prosecutors dealt with problems inherent in cases where most of the witnesses had died.

The killing happened in the dead of night. As the sun rose, a park worker found the woman, sprawled face down by the playground climbing blocks, her jeans pulled down and off one leg. He ran to find a telephone to call the police. When detectives turned her body over, they saw she was bleeding from her face. The Medical Examiner found three stab wounds that went through her cheek and into her mouth. The Cause of death was manual strangulation.

It was 1986 and there were over 1,900 homicides in New York City that year. The pathologist took body cavity swabs, clipped her fingernails, bagged her clothes, and went on to the next cadaver. Police investigated, but the case was closed with all leads exhausted. The murder of Antoinette Bennett was another case gone cold.

Law enforcement recently began looking to science to see whether new DNA technology could recover evidence in “up

close and personal” murders like this one. In 2010, the Cold Case Unit of the OCME’s Forensic Biology lab used a new technique to process fingernail clippings for DNA. Instead of swabbing the nails, criminalists put the whole clipping into a test tube and used a method called “sonication” that uses sound energy to literally shake the DNA off the nail and into the solution, where the cells get disrupted even before extraction.

Using this method, the criminalists developed a full male profile from those fingernails, which hit in CODIS to a 50-year-old man. As it turns out, Antoinette Bennett wasn’t his first victim. Steven Carter had raped, beaten, and choked his teacher into unconsciousness when he was only 16.

After detectives spoke to Carter in prison, prosecutors asked a new pathologist to examine old autopsy reports and photos, and cold case criminalists worked off lab reports completed decades earlier to bring this case out of the cold and into a courtroom.

Learn how prosecutors presented a 25-year-old homicide in a modern courtroom, when witnesses who did important work on the case were no longer available to testify. Watch interviews of the detectives, criminalists, and pathologist who testified at Steven Carter’s murder trial, and see the crime scene photos that were as compelling at trial in 2011 as they were the day they were taken.

DNA, Fingernails, Cold Case

E29 *Maryland vs. King*: Compulsory Extraction of DNA and the 4th Amendment

James K. Record, LL.M., California University of Pennsylvania, Dept of Justice, Law & Society, 250 University Avenue, California, PA 15419*

After attending this presentation, attendees will gain an understanding of the reasoning behind the majority and minority opinions in *Maryland vs. King* as well as the far reaching impact upon citizens and their constitutional rights under the Fourth Amendment.¹

This presentation will impact the forensic science community by pointing out that during the oral argument in *Maryland vs. King*, Justice Alito announced that he regarded King as “perhaps the most important criminal procedure case this Court has heard in decades.”² The holding espoused in *Maryland vs. King* impacts the forensic community, law practitioners, and society.

One of the most significant scientific advances made in law enforcement techniques is the development of the capacity to test an individual’s genetic markers and thus to learn that person’s identity. Because an individual’s DNA is entirely unique to that person, revealing that person’s genetic profile is solid evidence of who that person really is. Thus, DNA is a powerful tool to identify an individual by links to physical evidence obtained at a crime scene, and, alternatively, completely exonerate the individual if there is no match.

The U.S. Supreme Court in *Maryland vs. King* was asked to clarify the power of the police, under the Fourth Amendment, to take a DNA sample from a person accused of a crime. At issue was a Maryland law that required a routine DNA sample of every person arrested by the police for what the law defines as a “serious crime.”

The threshold question in *King*, as in many Fourth Amendment cases, is whether a search occurred? The issue was really not in dispute. As the majority opinion points out, cases under the “reasonable expectation of privacy” rubric of *Katz vs. United States* had held that requiring an individual to expel air from deep within his lungs, to submit to a blood draw, and to have

debris gently scrapped from beneath a fingernail all had been deemed searches.³ Scraping the inside of the cheek falls in this same category. Yet, a sharply divided Supreme Court upheld the power of the government at all levels to take DNA samples from every single person legally arrested for a “serious” new crime.

Justice Kennedy, writing for the majority, insisted that the ruling involved little more than what happens when the police take a suspect’s fingerprints or mug shot. Justice Scalia, writing for the dissent, said the Court validated the use of scientific evidence taken without a warrant not to make identification, but to gather evidence to solve cold cases — something he said the Court has never allowed before, thereby obliterating the long-standing rule that police may not take scientific samples from an individual, if the sole purpose is to solve a prior crime.

Ultimately, the Court found that the public policy value of a procedure of routine DNA sampling of arrested persons is so great, and DNA technology so efficient, that this far outweighs the minimal intrusion on privacy that is entailed when police take a DNA sample.

The testing of a person’s DNA simply because of an arrest implicates serious privacy concerns. It involves a bodily intrusion for an individual’s genetic blueprint, and the information it reveals is increasingly used for familial searching, thus extending its reach far beyond the actual person arrested.

Of critical importance is that neither the majority nor minority opinion opined about a major Fourth Amendment issue. What happens if after an arrestee is sampled and a match is made to an unsolved crime, the arrest is deemed to be unlawful? Does the Exclusionary Rule mandate that the information collected from the DNA extraction be excluded from any prosecution for the unsolved crime as a fruit of the poisonous tree?

In conclusion, although the extraction of DNA from an innocent person under the law may be minimally invasive as the majority insists, the minority makes a compelling argument that the Maryland law is in place, not for identity purposes, but for investigating unsolved crime unrelated to the original arrest. Furthermore, the holding albeit simple-to-understand contention, has created complex Fourth Amendment issues concerning DNA, privacy verses policy, and criminal procedure issues.

References:

1. 569 U.S. ____ (2013), 133 S.Ct. 594 (2013)
2. See Transcript of Oral Argument in *Maryland vs. King*, No. 12-207 (Feb. 26, 2013) at 34.
3. 389 U.S. 547 (1967)

Privacy, Arrest, Seizure

E30 Putting the Heat on Cold Cases: How to Identify and Evaluate Cold Cases — A Collaborative Approach

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After attending this presentation, attendees will learn how new DNA testing methods and grant funding enabled prosecutors to review hundreds of old unsolved homicides and the challenges encountered by law enforcement, district attorney’s offices, and the forensic laboratory during this process.

This presentation will impact the forensic science community by illustrating how to identify and evaluate potential cold cases for review and testing.

This presentation will help attendees learn how after receiving a National Institute of Justice (NIJ) Cold Case grant in 2010, Bronx County prosecutors were able to evaluate and review hundreds of cold case homicides looking for potential probative crime scene evidence to submit to the forensic biology laboratory

for DNA testing. The team of prosecutors and detectives developed guidelines for case selection and prioritization criteria to ensure the greatest likelihood of a DNA match and eventual prosecution. Such criteria helped investigators whittle down a potential pool of more than a thousand unsolved cases to a more manageable number.

The grant team has reviewed over a hundred files pertaining to unsolved homicides which occurred between 1990 and 2000. Specifically, the Bronx County prosecutors concentrated on *close contact* homicides involving strangulation, stabbings, or physical assaults which would yield the greatest chance of finding biological material suitable for forensic analysis and which would allow prosecutors to pursue new potential leads.

Prosecutors created an electronic database designed to organize all the police paperwork, forensic files, and photographs associated with the case. This allowed the prosecutors to track the progress of the grant, as well as easily find case documents when needed. Investigators from Bronx Homicide Task Force and The NYPD Cold Case Squad reinterviewed witnesses and researched possible suspects.

This presentation will also document some of the legal obstacles involved when working on these types of cold cases including locating witnesses, tracking down original files and police paperwork, and ultimately hunting down crime scene evidence which has been stored in police department warehouses for years, sometimes decades. Locating evidence has proven to be the largest hurdle following Hurricane Sandy in October 2012 which devastated parts of the New York region and resulted in the severe flooding of several New York City Police Department property warehouses.

Work conducted under this Cold Case grant has yielded some exciting results. In one such unsolved case, the Bronx County team requested that NYC Office of the Chief Medical Examiner, Forensic Biology laboratory retest the intimate body swabs from a 1998 cold case homicide that was included in a larger sexual assault pattern in Bronx County. Prior DNA testing revealed a partial male DNA profile, but technology at the time was not sensitive enough to develop a full profile suitable for inclusion in CODIS. Retesting of the swab using advanced, more sensitive technology was successful and a full male DNA profile was developed and uploaded to the New York State DNA Databank. Within months, the Bronx County District Attorney (BXDA) was notified of a forensic match between the male DNA profile from the postmortem sexual assault evidence kit and a convicted offender whose DNA profile was already on file from a prior DNA-qualifying conviction. This investigatory lead was thoroughly explored, including interviewing old witnesses and collecting police/OCME paperwork associated with the case. The identified suspect was eventually arrested and indicted on Second Degree Murder charges in March 2012. This success story illustrates the incredible impact that such projects can have and the potential for solving many more difficult, unsolved cases in the future.

DNA, Law Enforcement, Cold Case

E31 You Call That a Coincidence? The Significance of DNA Database Pairwise Comparison Searches in the Cold Hit Case

Scott M. Kozicki, JD, Office of Cook Co Public Defender, 69 W Washington, Chicago, IL 60602; and Brian J. Walsh, JD*, Office of Cook Co Public Defender, 69 W Washington, 17th Fl, Chicago, IL 60602*

After attending this presentation, attendees will be aware of the significance of litigation in producing the actual number

of arrestee- and offender-matches in the DNA database. This presentation will include how much more common DNA profiles are found to match in such databases in contrast to the statistical calculations presented by the government at trial.

This presentation will impact the forensic science community by discussing recent discovery litigation and disclosures which can enhance the reliability and credibility of forensic DNA statistical practices.

Illinois is the first state having a statute authorizing the court to order the production of this type of evidence. Though many other courts and labs have strenuously objected to production of this data, keeping such data hidden from the scientific community is fundamentally wrong. Scientists must openly share their data with one another to challenge all prior assumptions and test existing ones. To ignore this scientific principle by allowing government laboratories to hold onto data without the rigor of examination from outside scientists contradicts the universal principles of good science.

Undoubtedly, the existence of arrestee and offender databases has greatly assisted law enforcement with DNA leads for otherwise closed cases. The statistical calculations that are used showing the rarity of a DNA match have assisted prosecutors in securing countless convictions throughout the country.

But as the size of arrestee and offender databases increase, the likelihood of a coincidental match increases despite the great mathematical odds presented in court to the contrary. Though scientists and defendants have requested the actual data in these databases to examine their significance, the government has resisted all scientific inquiry, calling it not relevant and meaningless.

The actual data suggests otherwise. In 2005, an Arizona crime lab analyst ran a pairwise comparison analysis of the Arizona offender databank. At the time, the databank contained 65,493 profiles. The results showed that the databank contained 122 pairs of profiles matching at nine of 13 loci, 20 pairs of profiles that matched at 10, one pair that matched at 11, and one pair that matched at 12. The 11 loci pair and 12 loci pair were later found to be siblings.

In 2006, a database search of the Illinois Offender Database was ordered and the Illinois State Police initially determined that 903 pairs of nine loci match out of the initial search results released pursuant to defense requests indicate that 28 pairs had 11 loci in common with further loci that indicate the pair is not from the same person; five pairs had 12 loci in common; one of these pairs has further loci that indicate the pair is not from the same person; 1,936 pairs had 13 loci in common; 17 of these pairs have further loci that indicate the pair is not from the same person; 13,618 pairs had 14 loci in common. Of these pairs, 119 have further loci that indicate the pair is not from the same person; 41 pairs had 15 loci in common; and 678 pairs of specimen had 16 loci in common. It is noted that the results were qualified as not verified for twins and multiple profile entries for the same individual and court order verification of the results is pending. The recent results also combine profiles tested at different number of loci.

Disclosure of this data would enhance the reliability and credibility of forensic DNA statistical practices. This will become increasingly important as the National Database continues to grow in excess of 10 million entries.

DNA Database, CODIS, DNA Statistic

E32 The Constitutionality of the Cold Case CODIS Hit

Justin J. McShane, JD, 3601 Vartan Way 2nd Floor, Harrisburg, PA 17110*

After attending this presentation, attendees will be exposed to the scientific possibilities that modern DNA presents and the law.

This presentation will impact the forensic science community by continuing to seek, identify, and discuss the interesting intersection of the public need to solve crime, the scientific possibilities that modern DNA presents, and the law.

There is little doubt that as a crime-solving tool, the Combined DNA Index System (CODIS) is very helpful for law enforcement. However, there is great tension that increased technology affords us in the inventorying, logging, and indexing of genetic material versus privacy concerns. The Fourth Amendment of the United States Constitution presumes a warrantless and suspicionless search is invalid. This tenant of constitutional law has been sacrosanct since before the founding. Yet every day, the federal and state governments conduct thousands of searches of its CODIS without any level of individualized suspicion and absent a warrant or any judicial intervention. As of January 2013, the national CODIS database contained more than 11,977,900 profiles of not just offenders, but many others. Once in CODIS, these profiles remain even past death. When police search CODIS, they compare a profile generated from a crime scene sample against each of the millions of profiles that constitute the national offender index. If there is no "matching" entry, the unknown that is developed from the crime scene continues to be searched on a regular basis and subjected to the ever-expanding CODIS profile system. Under this modern computer-aided police dragnet, the information database "cold hit" identify potential suspects. The results from these searches are then used to form probable cause to develop search warrants to obtain exemplars from the putative "match." The search warrants are executed and the exemplar from the now-identified person is compared to the crime scene and the evidence developed then and there. With these "matches," a prosecution comes about. There is power in the statistical information that is reported out in these cases with the result often being a guilty verdict with little corroborating evidence independent of the "cold hit." But for the resorting of CODIS to aid in the prosecution, many of these "cold hit" cases would never have been "solved." Despite the ultimate holding, the briefs and the arguments during the United States Supreme Court's argument of *Maryland vs. King* has now cast into serious doubt the propriety of this practice of warrantless and suspicionless searches from the police-only database.¹

Reference:

1. *Maryland vs. King*, 569 U.S. ____ (2013), 133 S.Ct. 594 (2013)

DNA, CODIS, Cold Case

E33 DNA Mixtures in Criminal Litigation: Common Issues With DNA Mixtures at Trial

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After attending this presentation, attendees will understand some of the common issues that occur when DNA testing produces a mixture profile from an evidence sample.

This presentation will impact the forensic science community by educating attorneys and other "non-science"

individuals involved in criminal litigation about common issues with mixture DNA evidence.

Mixtures of DNA profiles found on evidence at crime scenes are becoming more and more common in criminal trials. As DNA testing has become common in criminal cases, with an explosion in DNA testing for property crimes, attorneys, judges, jurors, and other non-science individuals involved in criminal litigation have had to become familiar with the science involving DNA testing. Mixtures can be particularly difficult for a non-science person to understand. Mixtures frequently occur when common evidence items are tested, such as guns, objects dropped by an offender, clothing, car doors, steering wheels, window frames, and door handles. This presentation will use a specific case to highlight some issues that must be addressed such as major/minor calculations, partial profiles, drop-out, multiple runs at different injection levels, as well as statistics issues.

The case example is a real case is presented with all identifying information of the defendant, victim, and analyst removed. The case was a first-degree murder case tried in the Circuit Court of Cook County in January 2013. The victim was found dead in the passenger side of his car. A witness saw the car involved in an accident and then saw the driver leave the scene. When EMS arrived, it was discovered that the victim had a gunshot wound in his left side. The driver's side air bag was swabbed and sent to the Illinois State Police crime lab for testing. The analyst tested the sample on Profiler® Plus and COfiler®. The profile generated from the testing revealed a mixture of at least three individuals from which the victim was excluded. A major profile was deduced at eight loci by the analyst and a CODIS search was conducted. A match to the defendant led to his arrest and subsequent identification by the eyewitness. The defense used at trial did not involve a challenge to the DNA due to other evidence in the case. Therefore, this is not a war story. It is; however, an excellent example of a real DNA-evidence profile which shows common issues with mixtures.

The sample was run at 5- and 10-second injections on two machines. Data from different runs were relied on to deduce the profile. The profile also had a great deal of data at some loci and none at others, indicating that the sample was degraded or inhibited or both.

A major contributor was deduced from the mixture. RFU levels varied greatly between loci so the major contributor is clear at some loci but not so clear at others. This impacted the decision by the analyst as to which loci will be included in the CODIS search. It also impacted the decision of what statistical model to use, the RMP or the combined probability of inclusion. The great variance in RFU levels also means that the analyst had to deal with the issue of drop-out.

The purpose of this presentation is to educate attorneys (prosecution and defense) on how to spot issues when they review testing and so that they can be better prepared to discuss these issues with the testing analysts as well as any experts that might be retained. Additionally, it is to help the attorneys with handling these issues at trial when evidence is presented to the jury.

Mixtures, DNA Evidence, Criminal Litigation

E34 The Time Has Come to Analyze DNA Profile Databases

Dan Krane, PhD, 3640 Colonel Glenn Highway, Dept Bio Sci, Dayton, OH 45435; and Roger G. Koppl, PhD*, Syracuse University, Dept of Finance, 721 University Avenue, Syracuse, NY 13244*

After attending this presentation, attendees will have a better understanding of the need for disclosure of the National DNA

Index (NDIS) database.

This presentation will impact the forensic science community by increasing awareness of the questions that could be addressed by an independent, transparent analysis of the profiles contained in the NDIS database.

The Federal Bureau of Investigation (FBI) has controlled the NDIS since it was established in 1994. Even though the federal legislation that established NDIS explicitly anticipated that records within the database would be available for purposes of research and quality control, the FBI has published no research derived from NDIS and has vigorously declined to disclose these records to academic scholars for the past 20 years. The time has come for the FBI to release anonymized NDIS profiles to academic scientists.

Analyses of NDIS profiles would allow independent evaluation of some of the population-genetic assumptions used to generate statistical weights for DNA profile matches with samples large enough to allow sensitive evaluation of population structure. Databases that have been made available for such analyses at the present time are relatively small collections of hundreds of individuals while NDIS presently contains more than 10,000,000 complete 13-loci STR DNA profiles. The relative utility of local, state, and nationwide allele frequency databases could also be assessed for the very first time.

Other analyses that thus far have only been possible with simulations could finally be performed with real-world data. Those analyses include (but are not limited to) determination of: the frequency with which three-person mixtures actually appear to be two-person mixtures; the effect of close relatives in a database upon match probabilities, and; the extent to which identity by descent causes clustering of DNA profiles. In a different vein, quality assurance analyses of a government database from Victoria, Australia, containing 15,021 9-loci STR DNA profiles found an error rate of approximately 1 in 300 and raised concerns about missed opportunities to develop investigative leads that could easily apply to the NDIS database as well.

Disclosure of the 13-loci STR DNA profiles in NDIS would not trespass on privacy interests in any meaningful way. The U.S. Supreme Court's recent ruling allowing for the inclusion of arrestees DNA profiles to be entered into databases like NDIS in part relies upon the fact that the 13 STR loci that are genotyped are non-coding and, therefore, cannot reveal sensitive information. Copying of the NDIS database itself should not be overly burdensome and would probably be accomplished in a matter of minutes.

Open access to data is a fundamental tenet of science. The need for openness in forensic science in particular was reinforced by a National Research Council report five years ago that decried the insularity of forensic science and called for greater involvement of the academic community in assessment, validation, and improvement of forensic science methods. The FBI should honor the norms of science and open the NDIS database to independent scientific scrutiny. Doing so poses no meaningful risk and can only strengthen the quality of forensic DNA analysis.

Database, DNA, Quality Control

E35 The Discovery Motion for "Scientific Stuff:" Don't Expect to Get It, Find It, or Recognize It (Even if You Do Get It) if You Have No Idea What You're Looking For!

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The goal of this presentation is to provide a lawyer with the answer to the question: What am I looking for and what could it mean? This presentation focuses on documentation which should

be available through a discovery motion for review by both sides as a part of pre-trial discovery. Specific technical documents and operational protocols will be identified.

This presentation will impact the forensic science community by providing lawyers with an enhanced knowledge base of what to request in a discovery motion and what to expect from the forensic science laboratory. Since the credibility of a forensic scientist's testimony could be inversely proportional to the number of four-syllable words which are used without an explanation in a "lawyerly discussion," the technical jargon in the presentation will be minimized. There are required laboratory documents which the defense should request and which the prosecution should be aware of so that both sides can fulfill their responsibilities to courts in evaluating the elements associated with the case at hand.

Some of the more common questions lawyers wonder about in reviewing a forensic analysis or discussing the analysis with the "expert" are:

- What does "that" mean?
- Why didn't someone tell us about this?
- What do all those big words mean?
- How am I supposed to cross-examine an expert when I have no idea what he/she is talking about or when I don't understand what was done in my case?
- Standards, what are standards?

Most forensic science laboratories in the United States have nothing to hide and issue reports which reflect conformance to standards. Most laboratories have subscribed to the oversight of external accrediting bodies and statewide commissions to maximize the probability that everything is transparent in the management and operational aspects of valid science. Disclosure of what is happening behind those doors with signs dictating "Authorized Personnel Only" is there for the asking; however, there are times when the defense and even the prosecution should be more inquisitive in seeking answers to questions which have a legal impact on what is happening behind those locked doors. The prosecution can usually pay a visit to the laboratory for a "sit down." The reality is that these "sit downs" with the prosecution occur far too infrequently. The "other side" faces a more daunting challenge. The defense, if they want to see anything, may be asked to get a court order. Sometimes there may be valid reasons for denying admission to certain parts of the laboratory (like the vault) to "unauthorized personnel." Even when the defense is allowed into the laboratory, all they will usually see are white coats, dropper bottles, flasks, microscopes, humming machines with robotic arms picking up small rubber-capped vials, and row after row of computer monitors with images of straight lines, curved lines, or columns of numbers.

The properly worded discovery motion is one effective way to obtain the answers to the most important questions. The challenge for the lawyer is this: how do you get what you need if you don't know what you are looking for? And if "you do get it," what does it all mean? Many discovery motions are couched in terms of: give me everything you've got. Even if and when "everything" is provided, the lawyer probably has no idea what "everything" (all that paper with charts and lines and paragraphs with four to six syllable words) really means.

This presentation will identify case-specific documents related to laboratory operations. Those factors which are important in determining the reliability of any scientific analysis conducted in a laboratory can be "discovered" by evaluating this documentation related to how the laboratory is managed: whether the laboratory is conforming to scientific standards which have become pro-forma requirements in forensic science laboratories; whether the data in the case at hand conforms to reporting protocols; whether all of the documentation supports the conclusions; whether alternative explanations are possible for those conclusions; and, whether all of the information which might be exculpatory is being provided.

A properly worded discovery motion for specific documents can provide some level of assurance that the answers to the relevant questions related to the case at hand are actually being provided, remembering that nothing is absolute. Standards do exist and conformance to those standards is the foundation of a laboratory's credibility.

Discovery Motion, Scientific Standards, Scientific Data

E36 The Era of Designer Drugs: An Ever-Changing Landscape

Heather L. Harris, MFS, JD, 2401 Lombard Street, Ste 1, Philadelphia, PA 19146*

After attending this presentation, attendees will be familiar with: (1) the legal and scientific classifications of designer drugs; (2) the variety of legislative approaches to designer drugs, including class control, and analogue statutes; (3) the issue of designer drugs in sentencing; and, (4) recent legislative proposals from Congress and the states.

This presentation will impact the forensic science community by educating attorneys in the changing areas of controlled substance law, thus improving their advocacy skills in the courtroom.

Over the past 50 years, a relatively small group of drugs, including marijuana, cocaine, amphetamines, heroin, and a handful of diverted prescription medications, such as opiates and sedatives, made up the vast majority of drug possession and distribution cases. These substances were easily recognized by the police and easily identified by the crime laboratories. Designer drugs, which are designed to thwart both of those layers of law enforcement, first appeared in the late 1970s and has remained mostly in small, isolated communities of drug users for 35 years.

However, around 2009 to 2010, the illicit drug markets began to see new compounds. Entrepreneurial chemists with access to global shipping networks began to synthesize compounds previously hidden away in scientific journals and patents and distribute those compounds through Europe and then into the United States. These new designer drugs came in two main categories: stimulant and/or hallucinogenic drugs, such as "bath salts" or "plant food;" and, synthetic cannabinoids, compounds intended to work like marijuana but often with some very different effects. The synthetic cannabinoids, sold as "Spice" or "K2" most famously, represented an entirely new class of compounds in the world of controlled substance law enforcement.

Initially, the legislative control of the bath salt compounds utilized traditional mechanisms, such as acts of Congress or administrative scheduling. When compounds appeared that were not controlled, the standard analogue statutes provided another mechanism for prosecution of these cases. Recently, some states are trying new legislative approaches to these compounds in order to avoid difficult analogue prosecutions.

On the other hand, new synthetic cannabinoids came onto the market at such a rush in 2010 and 2011 that it was quickly apparent that the traditional means of scheduling drugs could not keep up with the marketplace. Out of this dilemma, class legislation was born. This represented a significant change from the prior 40 years of controlled substance law.

One of the major challenges for attorneys with class legislation is that more chemistry is involved in proving the major element of the crime: that the alleged compound is within the controlled class. This presents a challenge for prosecutors who need to prove this element to a jury, and this could be an opportunity for defense attorneys who can now challenge the classification of a compound that is not specifically listed in the statute.

When neither traditional scheduling nor class legislation includes a new designer drug that has been identified in the market, the prosecutor has the option of an analogue prosecution. While these cases have traditionally favored the prosecution, recent rulings indicate a shifting landscape in this mature, yet relatively unnoticed area of the law. Today these cases offer challenges and opportunities to both parties.

Because of the ever-changing market of designer drugs, legislatures across the country are trying new approaches to controlling today the drugs of tomorrow. To illustrate the challenges in this new landscape, this presentation will include some specific examples of emerging designer drugs for attendees to assess their legal status as controlled substances or analogues.

Designer Drug, Legislation, Analogue

E37 Measuring Nothing: Limitations of Quantitative Confirmation Analysis and the National Trend Toward Zero Tolerant Per Se THC Limits in Drivers

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After attending this presentation, attendees will appreciate the need for accurate and precise toxicology confirmation analysis in citizens accused of driving with the presence of various levels or under the influence of marijuana.

This presentation will impact the forensic science community by highlighting how confirmatory testing and its limitations, both qualitatively and quantitatively, need to be understood in the forensic community. These limitations will be illustrated by real-life examples of the unjust consequences which can occur when reliable methods for analysis and reporting are not followed.

Many states have enacted laws making it illegal to drive with any amount of marijuana or its metabolites in the body. The recent trend to equate guilt based on either per se levels or even zero tolerance limits for marijuana in the body is based on incorrect assumptions. Those assumptions are that a driver is actually impaired and unsafe to other drivers on the roadway based on the presence of a certain amount or even "any amount" in the driver's blood. Under per se marijuana laws, a driver may be convicted even when a driver is not actually impaired as a result of ingesting marijuana. This raises two issues: (1) does science support that all drivers are impaired at a certain level?; and, (2) can these low or zero per se levels actually be determined with scientific reliability?

To complicate matters, eighteen states reformed or are attempting to reform marijuana laws through new legislation or through the initiative process (like California in 1996 and Colorado in 2012) to make ingestion of THC legal so long as there is a therapeutic basis as determined by a licensed physician. Most recently, the citizens of the states of Colorado and Washington voted to legalize marijuana consumption in persons over the age of 21 with no therapeutic reason nor any physician supervision required.

The Executive Branch of the Federal Government in its 2012 National Drug Control Strategy Report recommends the imposition of zero tolerant per se limits for marijuana across the country despite various state's law.¹ As of the date of this abstract, ten states (Arizona, Georgia, Indiana, Delaware, Iowa, Rhode Island, Wisconsin, Utah, Michigan, and Oklahoma) have enacted

zero tolerance laws for cannabis, with Oklahoma being the most recent. Yet, only Arizona and Rhode Island have made laws to exempt medical marijuana patients from prosecution under these per se statutes unless actual impairment is proven. The Michigan Supreme Court followed the logic of Arizona and Rhode Island law makers and authored a unanimous decision *People vs. Koon*.² In *Koon*, the Court held that drivers who fall under the Michigan Medical Marijuana Act may not be criminally convicted of being "under the influence" unless evidence is available that the driver is under the influence with reliable testimony.³

If citizens are being prosecuted for driving under the influence of marijuana based on toxicological analysis, then the science used to convict must be reliable. Limits of detection and limits of quantification remain at the crux of fair per se THC laws. Validated methods including the analysis of systematic and random uncertainties, are required to express a positive result with reasonable confidence. Absent reliable proven methods, we cannot be confident that reporting a THC amount is scientifically reliable.

References:

1. http://www.whitehouse.gov/sites/default/files/ondcp/2012_ndcs.pdf
2. Docket No. 145259, Decided May 21, 2013
3. MCL 333.26421 et seq.

Per Se THC Limits, Zero Tolerance, Unconfirmed Results

E38 How State Opinion Evidence Utilizing Unvalidated Drug Recognition Evaluations Can Mislead Juries

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The goals of this presentation are to provide attendees with: (1) an overview of the use by law enforcement of the Drug Examination and Classification Program (DECP) in America; (2) a summary of why reporting a drug concentration can be misleading; and, (3) why even under the more inclusive *Frye* standard a hearing is required to prevent the introduction of unreliable evidence.

This presentation will impact the forensic science community by examining the ways in which unvalidated drug recognition evaluations influence jurors.

The Unvalidated and Uncorrelated Drug Recognition Evaluation, where the certified Drug Recognition Examiner/Officer (DRE) is correct only 28% of the time about what drug is causing impairment in the arrested Driving Under the Influence (DUI) drugged driver, is perfectly acceptable, according to the International Association of Chiefs of Police (IACP). The previous sentence sounds incredulous, yet it is completely true.

The DECP program claims that upon completion of the 72-hour training regimen, the officer can diagnose: (1) if the suspect is impaired such that he/she cannot operate a motor vehicle safely; (2) if that impairment is caused by drugs; and, (3) which of the seven drug classifications is causing the impairment. DRE officers claim the ability to do this by administering the twelve-step battery of subjective testing.

The gatekeeper function of the court, as applied, requires that before the opinion of a DRE officer who conducted the evaluation can be introduced in court, a foundational hearing should be held to determine whether the opinion passes muster. The defense right to *voir dire* the "expert" regarding the opinion he/she intends to offer is proper and well established yet routinely is

overlooked.

The National Academy of Sciences agrees and has recommended: "The degree of science in a forensic science method may have an important bearing on the reliability of forensic evidence in criminal cases. There are two very important questions that should underlie the law's admission of and reliance upon forensic evidence in criminal trials: (1) the extent to which a particular forensic scientific methodology that gives it the capacity to accurately analyze evidence and report findings; and, (2) the extent to which practitioners in a particular forensic discipline rely on human interpretations that could be tainted by error, the threat of bias, or the absence of sound operational procedures and robust performance standards. These questions are significant. The goal of law enforcement actions is to identify those who have committed crimes and to prevent the criminal justice system from erroneously convicting the innocent. So it matters a great deal whether an expert is sufficiently reliable to merit a fact finder's reliance on the truth that it purports to support." The forum in which to determine the reliability of expert opinion is in a *Frye* or *Daubert* hearing and occurs before the opinion is allowed as evidence.

The DECP program does not comply with the criteria for reliable testing as set forth in the Consensus Report addressing drug concentrations and impaired driving published in the *Journal of the American Medical Association*. Even the most intelligent officers who successfully complete the DRCP program are not equipped to understand how drugs interact in the body. The DRCP program mistakenly believes that 37 hours of substantive classroom lecture is adequate to educate the DRE officer in both the pharmacokinetic effects and pharmacodynamic effects of seven distinct drug classifications.

In order to accurately determine if a suspect is impaired by drugs, the officer must be educated in the interactions of the chemicals found in the drugs with the biological receptors in the brain (pharmacodynamics). The DRE officer must also understand fully how the chemicals from the drugs are absorbed, distributed, and eliminated in the body (pharmacokinetics) and how long that process takes.

The reliability of the DECP program is clearly flawed. Officers lacking any scientific or medical background are admitted into Drug Recognition "Expert" school and later certified after minimal training. Despite the fact that it has been shown that officers' predictions of actual drug classification are commonly incorrect, decertification for poor performance including misidentification is rare, at best. Each day judges continue to allow unreliable DRE testimony to be introduced to juries who in turn have little choice but to convict based on unvalidated DRE opinion evidence cloaked in the aura of science.

Drug Recognition Examination, Unvalidated Opinion, Incorrect Drug Classification

E39 Widmark's Alcohol Equation: A Forensic Application for Litigation

Jay Zager, 10638 NW 69 Street, Parkland, FL 33076; and Mary C. McMurray, BS*, 3523 County Road JG, Blue Mounds, WI 53517*

After attending this presentation, attendees will understand the simplified forensic application of conducting Widmark's Alcohol Equation in Driving Under the Influence (DUI) and other alcohol investigative-related litigation.

This presentation will impact the forensic science community by reinforcing the necessity of properly applying Widmark's formula to address assumptions and avoid false and misleading results in judicial proceedings.

Widmark's Equation is an algebraic formula first published

by Dr. Eric M.P. Widmark in 1932.¹ The equation is frequently utilized in courtrooms to determine the total number of alcoholic drinks in the human body based upon a forensic breath, blood or urine alcohol sample.² The equation has also been used in an attempt to perform a retrograde extrapolation of a measured alcohol concentration to a different point in time, typically the drive time. The latter application has been called "forensically unreliable" due to the number of unknowns including: absorption vs. elimination phase; peak concentration; elimination and absorption rates; and, short-term fluctuations in concentration.³

Widmark's Equation is often expressed as follows:

$$AC = \frac{A \times 0.8}{W \times R} \times 100$$

A = Mls of pure ethanol, 200 proof
AC = Alcohol Concentration, Chemical Test
W = Body weight in grams
R = Water: tissue volume of distribution: 0.55 for females; 0.68 for males
0.8 = Specific gravity of ethanol

While many versions of Widmark's Equation are used in courtrooms and have undergone many updates, a simplified version that combines the constants and conversion factors is expressed as follows:^{4,5}

$$AC = \frac{D \times 2.6}{P \times R}$$

Where:

P = Weight in pounds; 1 pound = 454 grams
D = Number of drinks; 1 drink = 1 oz. of 100 proof = 30 mls of 100 proof = 12 oz. of beer (~4.2% ABV) = 4 oz. wine (12.5% ABV)

For Alcohol Concentrations (AC) other than 100 proof, multiply the AC calculated for that drink (D) or portion of a beverage by the alcohol proof expressed as a percentage (e.g., for 80 proof alcohol, the user would multiply the AC by 0.80) Assumptions pertaining to the height/weight distribution and alcohol content of each drink need to be clearly stated.

This simplified version does not change Widmark's formula. It merely converts the formula into a manageable equation for the non-scientist by defining a "drink" as beer, wine, or spirits and incorporating U.S. Standard Units of pounds, alcohol proof, and fluid ounces into the formula.

Historically, Widmark's formula did not include an estimate of uncertainty. The equation includes at least seven random uncertain variables, thereby having significant uncertainty. Failure to address and report appropriate uncertainty estimates when applying Widmark's Equation decreases its confidence and acceptability in scientific and legal contexts.⁶

Calculations other than estimating alcohol concentration or number of drinks require additional information; for example, a person's absorption rate for each drink and elimination rate. These calculations introduce additional uncertainty to the estimates of drinks and alcohol content and must be taken into consideration.⁶

Commercial software is available for performing the Widmark Equation and calculating associated uncertainty with varying interpretations of case facts, including alcohol concentration throughout the drinking session and beyond.⁷ Many of these programs will generate graphs and tables for use as demonstrative aids for litigation.

When relying on Widmark calculations, especially

with commercial software, it is important to avoid expanding or embellishing facts and known constants simply because the program can incorporate varying scenarios. According to Dr. Kurt Dubowski, "No forensically valid forward or backward extrapolation of blood or breath alcohol concentrations is ordinarily possible in a given subject and occasion solely on the basis of time and individual analysis results."⁸

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Widmark Alcohol Equation, DUI, Blood Alcohol Concentration

E40 The Measurand Problem in Breath Alcohol Testing

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After attending this presentation, attendees will have a better understanding of: (1) how the law and science interact to define the measurand of a breath test; (2) where and how this interaction has created confusion, what forensic breath alcohol tests actually measure and their results mean; and, (3) what factors should be considered in the determination of the uncertainty of breath test results.

This presentation will impact the forensic science community by dispelling a source of confusion that often leads to breath alcohol test results being misinterpreted by those presenting and relying upon them. Attendees will be better able to present breath test results for what they actually represent, dispel erroneous legal challenges, and determine the uncertainty associated with breath test results. This should facilitate enactment of per se legislation that avoids the measurand problem and the establishment of a nationally standardized breath test measurand for per se legislation.

Breath alcohol testing is relied upon to measure the concentration of alcohol in breath and, indirectly, in blood. The concentration sought constitutes the "quantity intended to be

measured," referred to as the measurand.¹ The measurand of a breath test is dictated by law and varies between jurisdictions. Thus, identical numerical values obtained from tests in disparate jurisdictions may refer to different quantities and may not indicate the relevant statutory quantity. This can lead to misinterpretation of results, referred to as the "measurand problem."

The measurand problem arises where the identity of the quantity subject to measurement and the quantity intended to be measured are distinct but not well specified. It is a common source of confusion in forensic breath alcohol testing. The origin of the measurand problem in forensic breath alcohol analysis is the fact that what constitutes an individual's alcohol concentration for purposes of a per se DUI offense is defined by law-making authorities, not science. To illustrate the confusion, the work of both Hlastala and Gullberg will be considered.^{2,3}

Hlastala's paradigm describes the dynamic processes that cause the concentration of alcohol in a sample of breath to continuously change as it is inhaled and exhaled. The mechanisms described determine what an individual's Breath Alcohol Concentration (BrAC) will be. Scientifically, Hlastala's paradigm is critical when considering the meaning and accuracy of breath alcohol test results. In the context of statutory/regulatory per se prohibitions; however, it is not science that determines the measurand of a breath test but the law. In many jurisdictions, Hlastala's paradigm is critical when considering the meaning and accuracy of breath alcohol test results. Many jurisdictions; however, define BrAC such that it is irrelevant for these purposes. The failure of forensic and legal professionals to appreciate this leads to reliance upon Hlastala's where it is irrelevant and its preclusion where it is relevant and critical.

As breath test programs have sought to become compliant with international standards, the uncertainty associated with breath test results has become a growing topic of discussion and confusion. Many have turned to the work done by Gullberg to help make sense in this area. Gullberg's methodology does not apply to the results of all breath tests; however, rather, it directly applies only to results obtained in a particular type of jurisdiction, the type where Hlastala's work is irrelevant. Gullberg's work can be extended to other jurisdictions under certain circumstances. This requires an understanding of what it is originally intended to apply to. This understanding evades many forensic and legal professionals.

This presentation will set forth three models of distinct but representative jurisdictional types defined by the manner in which per se statutes/regulations determine the measurand of a breath test. It will be shown how these laws operate to dictate not only what is measured and what BrAC results actually indicate, but how and where both Hlastala's and Gullberg's work does and does not apply. This will reveal the nature and impact of the measurand problem and suggest solutions that can be adopted to address it.

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Measurand, Uncertainty, Breath Testing

E41 Educating the Lawyer: The Birth of the Lawyer-Scientist

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The goal of this presentation is to help highlight the need for practicing lawyers to become educated in the scientific principles involved in their area of practice with a focus on drug- and alcohol-related driving cases.

This presentation will impact the forensic science community by encouraging education for lawyers in the areas of forensic science in the face of a history of the legal community running scared of science.

Many areas of law are increasingly becoming more and more scientific. Alcohol-related driving cases serve as a great example of this trend. More than ever before, blood is being drawn and tested for use as evidence in criminal trials. Historically, blood has only been used for testing, to any great extent, in suspected impaired driving fatality accidents. That is no longer the case. Many states have implemented or are in the planning stages to implement "no refusal" policies where a motorist suspected of driving under the influence will be forced to give blood for testing even when they refuse consent to testing. Blood testing is also on the increase because of the increased law enforcement awareness of drugged driving where a breath test is of no value.

For decades, the mantra of criminal defense lawyers has been, "when the law is on your side, argue the law — when the facts are on your side, argue the facts — when you have neither, muddy the waters." With this increased reliance on science in DUI/DWI cases, lawyers can no longer follow this mantra. They must learn the scientific principles behind the area of law they practice. Courses focusing on science instead of law are popping up across the country for attorneys to attend and learn directly about the science. This training is a must for lawyers who handle these types of cases. This presentation will focus on the importance of lawyers becoming trained in the sciences and discuss some of the resources available to lawyers to learn the science. The days of the jack-of-all-trades lawyer who handles any and every case that comes through the door are past. In today's legal world, lawyers must reinvent themselves and embrace science. They must join the ranks of the new breed of lawyer called the lawyer-scientist.

This need to reinvent and reevaluate is a duty of lawyers on both sides of the criminal justice system. It is just as important for prosecutors who are tasked with prosecuting cases ethically and fairly using legitimate scientific principles as well as a duty for defense lawyers to scrutinize and challenge forensic results.

Training Lawyers, Lawyer-Scientist, Science Education

E42 Crash Reconstruction Basics for the Legal Practitioner

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After attending this presentation, attendees will understand some basic concepts utilized in the field of crash reconstruction. The foundation of these concepts, along with some of the common myths and misuse, will be explored. Examples of each concept will be provided, along with a discussion of the importance of obtaining the underlying crash reconstruction file and what can be expected to be contained in the file.

This presentation will impact the forensic science community by familiarizing legal practitioners in both the criminal

and civil realm with basic crash reconstruction principles, helping them to more effectively communicate with potential experts the attorneys may choose to hire, as well as spot problems with crash reconstructions completed in their cases.

After being appointed as counsel on two criminal vehicular homicide cases in which the crash reconstructions lacked any foundation for the opinions offered by the State's reconstructionist, the counsel embarked on an educational mission in crash reconstruction. Prior to this endeavor, the only vehicle-related education that could be claimed was one year as a FedEx driver in Green Bay, Wisconsin.

Concepts to be explored in this presentation include friction, often referred to as "drag factor," including methods used to determine a road's drag factor, the effect of drag factors on speed analysis and whether temperature can affect the friction value of a surface. Two common methodologies used to determine traveling speeds for vehicles involved in a collision will also be discussed: (1) conservation of linear momentum; and; (2) critical speed analysis. The foundations for each method will be explained and their potential for misuse will be examined. The question of whether headlights and taillights can be examined to determine if they were on at the time of an accident will be discussed, along with the limitations of this evidence. Included in this will be an explanation of why a complete vehicle inspection is vital to a thorough crash reconstruction. Finally, this study will provide an overview of what an attorney can expect to find in a crash reconstruction file and discuss the importance of obtaining the underlying file and meeting with the opposing expert prior to trial.

Actual case file examples will be presented of each concept to better illustrate the principles so even attorneys with no prior crash reconstruction experience will have a basic understanding of what to look for in future cases.

Crash Reconstruction, Basic Concepts, Attorneys

E43 Particle Combination Analysis: A Fundamentally New Investigative Approach

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After attending this presentation, attendees will understand the distinction, contribution, and requirements of particle combination analysis as compared to more commonly used approaches in forensic investigations.

This presentation will impact the forensic science community by allowing program managers and policy makers to recognize the potential contributions of particle combination analysis and to understand the requirements for implementation using existing resources. This will lead to the exploitation of an extraordinarily useful form of physical evidence that is virtually ignored by current forensic science practices.

Particle combination analysis is a new approach that uses co-occurring particles to test alternative attribution hypotheses. Simply put, particle combination analysis exploits the particles in dusts, which are ubiquitous and in infinitely varying combinations, to solve a wide range of problems with varying case specifics. This approach can provide a game-changing capability to forensic investigators, working alongside existing investigative methods and using portions of evidence that are typically discarded or ignored.

The approach utilizes existing staff and laboratory resources in a fundamentally different way; it does not require major investments in training, equipment, or retention of outside experts. It does require a change to the way existing resources

are used.

An INTERPOL case involving contraband elephant ivory will be used to contrast approaches currently used in most forensic investigations and show how and why a different approach resulted in significantly better results. The same approach can make significant contributions to a wide range of investigations and cases.

Conventional approaches to forensic investigation include: (1) direct comparison to a specific suspected source; (2) classification using a reference library of potential sources; (3) application of a predetermined analytical tool; and, (4) analysis of specific components of the specimen. Each of these approaches can be useful and makes an important contribution, but each also has requirements that restrict its applicability, none of them fully exploits the available specimen, and their contributions are only serendipitously sufficient to address the needs of any particular investigation.

In contrast, particle combination analysis has no requirement of comparative analysis, no predetermined restriction on the type of analytical tool, and no predetermined restriction on the components within the specimen that are exploited. Rather, standing protocols are used for specimen assessment. The results determine the range of potential contributions to the resolution of case questions. Specific specimen components are analyzed using modular validated protocols that are not preselected, but are strategically chosen based on their potential to resolve case questions.

Using particle combination analysis, dusts from within a shipment of contraband ivory were analyzed to help determine the original location where the ivory was packed. Key findings were the types of minerals, soil, and vegetation represented in the dust, as determined using a combination of light and electron microscopy, energy dispersive X-ray analysis, infrared microspectroscopy, palynology, and non-human DNA analysis. Beginning with a possible origin within the continent of Africa, first-stage analysis of the recovered dusts was able to eliminate environments comprising approximately 91% of the area, including all areas of 36 countries. Of the remaining 12 countries, the analysis was able to eliminate 72% of their area, allowing the investigations to be focused within portions of these countries. Next steps were defined to further reduce the possible origins of the dust based on more detailed regional analyses.

The particle combination analysis approach resulted in useful information arising from many different particle types. Different combinations of particles resulted in environmental, geographical, and land use “signals” that addressed specific investigative questions in this case. Particle combination analysis is a general capability: it is not restricted to questions of geographical origin, to African elephants (or any type of wildlife), to sealed crated shipments, or to specimens containing any specific type of particles. Rather, it is an approach that extracts case-relevant information (for virtually any type of case) from the complex assemblage of particles that are found (on virtually any specimen).

Requirements for successful implementation using existing staff and resources include administrative protocols driving the facile use of multiple disciplines, incorporation of a case-level scientific investigative methodology, and parallel adjustment of institutional norms.

Augmentation of existing practices with this new capability will result in major contributions to many case investigative problems. The primary challenge is institutional willingness to use existing staff and equipment in a different way.

Particle Combination Analysis, Trace Evidence, Dusts

E44 The English Law of Witchcraft: Past, Present, and an Uncertain Future

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After attending this presentation, attendees will have an understanding of the evolution of the law in response to the changing mores of society as demonstrated by the law relating to witchcraft.

This presentation will impact the forensic science community by illustrating the adaptability of law through the actions of the legislature, judiciary, and international law, while retaining the capacity to prosecute harmful practices falling under the rubric of “witchcraft.”

While witchcraft has been defined as “the practice of attempting to harm people by supernatural means, but through emotions and thought alone, not through the use of tangible objects;” this is a narrow definition.¹ Roman law differentiated benign magic (fertility, finding treasure, divination, etc.) from malign magic (e.g., souring milk, blighting crops, etc.) with satanic magic as a category of malign magic. In pagan societies, where the perception was that most things were imbued with supernatural entities susceptible to entreaties by prayer and sacrifice, this was understandable. The monotheistic religions all preach against the practice of witchcraft. The best known injunction is, “*Thou shall not suffer a witch to live,*” but there are others.² The early Church had clear objections to sorcery.³ In medieval England, both the secular and the ecclesiastical courts tried witchcraft cases. The ecclesiastical courts treated the offence as a variation of heresy, with the degree of intent an important factor in determining guilt and punishment. Repentance led to reconciliation with the Church, with the death penalty being reserved for the unrepentant. The secular courts took a narrower view. Intent was not an issue and the sentence upon conviction was death. There was no “Burning Time” in England. Witches were executed by hanging.

Following the Henrician reforms, the Crown needed to exert control over the spiritual life of the population. One tool was the Witchcraft Act of 1542.⁴ This brought the prosecution of witchcraft into statute law. By the end of the 16th-century skeptics were beginning to be heard.⁵ However, witchcraft prosecutions continued into the 17th-century with the last English executions for witchcraft in 1662. All English witchcraft statutes were repealed by the Witchcraft Act of 1735. This replaced the offense of practicing witchcraft with the offense of purporting to practice witchcraft. In short, witchcraft was fraudulent and practicing it was a form of fraud. Toward the end of its life, the Witchcraft Act was used to prosecute mediums and spiritualists, with the last such prosecutions taking place in 1944. These prosecutions resulted from mediums offering to contact the war dead. The Act was repealed by the Fraudulent Mediums Act of 1951 that, in effect, legitimized Spiritualist Churches. That act in turn was repealed in 1993. Protection against fraudulent treasure finders and their ilk is the remit of consumer protection legislation.

The English law has moved from being neutral about witchcraft to being tolerant of it and even to supporting the rights of adherents to practice their religion. In 1977, a Court declined to recognize Wicca as a religion. In 1998, The Human Rights Act incorporated a substantial proportion of the European Convention on Human Rights (ECHR) into English statute law. With some limitations, the ECHR gives the citizens of signatory states the right to freedom of religion. Prisoners, police officers, and naval personnel are now allowed to profess adherence to Wicca and other neo-pagan religions. The Human Tissue Act 2004 exempted the use of human material used as relics in acts of public worship

from the licensing provisions the act applies to other lawful uses of human material.

In short, 21st-century English law is now silent on specific offenses relating to the practice of witchcraft. Practitioners of witchcraft are not exempt from the general criminal law and some interesting prosecutions have arisen as a result.

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Witchcraft, Jurisprudence, Society

E45 Medical Malpractice Cases and the Physicians' Rights in Turkey

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After attending this presentation, attendees will gain an understanding about the legal and ethical aspects of the patients' and physicians' rights and will start to raise awareness in the title question of "What about the Physicians' Rights?"

This presentation will impact the forensic science community by raising awareness of the physicians' rights as well as the patients' rights and discussing legislative problems in Turkey. Also discussed will be the distinctions between countries around the world on this issue.

World Health Organization (WHO) describes "health" as "a state of complete physical, mental, and social well-being and not merely the absence of disease or infirmity. The right of health is described as the rights for "the establishment and development of health." The Turkish legal system has new definitions in terms of civil and penal liability of medical doctors. The Turkish medical doctors are wishing for a reduction of financial burdens due to malpractice insurance and express the discomfort they feel from being frequently sued.¹

Because of the problems encountered during health services, fast and permanent solutions have been investigated in the last couple of years. Some legal arrangements have been made for more effective evaluation of health care personnel who made mistakes and new measures have been noted. Patients' rights have been protected with various legislations, arrangements, and regulations, yet health care personnel have still remained far behind all of these. As a result of the new regulations with aggravated circumstances, medical doctors face heavier penalties and indemnifications. Because of that, conditions today show us that none of the medical doctors may remove themselves from medical law suit cases. Even if all the unwanted consequences and complications, including the undefined ones, are avoided, the

payment of compensation due to malpractice cannot be avoided. Results of epidemiological studies show that only 2% of the real medical mistakes are reported to the courts. More striking is data showing that only 20% of the cases that end up at the courts are actually medical malpractice. This means that the medical defense system which is described as a kind of forensic consultation has to cope with an 80% portion using a serious medicolegal risk management.¹⁻⁷

The most important document on the subject is the "Declaration of Wrong Medical Practice (Malpractice)" adopted in the 44th General Assembly of the World Medical Association in 1992. According to this declaration, there should not be any obstacles to meet the loss of patients who suffered as a result of malpractice. Appropriate policies should be developed for medical doctors who have inadequate knowledge and skills, and medical doctors should be prevented from medical applications until their failure is eliminated.

The Organization for Economic Cooperation and Development (OECD) report on the "Medical Malpractice Insurance Coverage of Countries, 2005" shows that malpractice lawsuits are increasing every day and trust between the doctor and patient is being lost. Different applications are seen in each country. For example, in Belgium the "Order of Medical Doctors" which represents medical doctors, has been given the power of canceling the license and registry of the medical doctor who commits malpractice. In Portugal, the Portuguese Medical Association also has this authority. Malpractice issues in Denmark are resolved by The National Board of Health. Similar applications are in force in Hungary; however, deficient medical doctors are trained free of charge. The United States, United Kingdom, Sweden, France, and Finland have medical malpractice laws and practices similar to each other. Turkey has basic legal regulations.¹

Under Turkish law, in order to describe the act of a medical doctor as a crime, the action should be contrary to law, contain an element of the defect, result in damage, and there should be a relation between the damage and defect. As there is not a medical malpractice law in Turkey, the medical doctors are judged under general criminal and civil law. The Turkish Medical Association is involved in ethical aspects only. The penalties applied to medical doctors and other medical staff in case of faulty medical applications varies from one country to another. A special arrangement covering all countries does not exist for malpractice.¹⁻⁷

In this presentation, various concrete case examples will be examined and discussed according to legal regulations and, at the end, participants will have detailed information on the concept of malpractice in Turkey and universally, as well as the new regulations and the legal dimensions which need to be renewed.

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Medical Malpractice, Physicians' Rights, Patients' Rights

E46 An Inter/Multidisciplinary Approach of Münchaussen by Proxy Syndrome: The Role of Public Health Professionals, Courts, and Forensic Experts

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After attending this presentation, attendees will be able to discuss the role of public health professionals, courts, and forensic experts in cases of intrafamilial violence, especially concerning Münchaussen by Proxy Syndrome.

This presentation will impact the forensic science community by showing how public health professionals can contribute by providing evidence and technical information firsthand in order to prevent crime and protect potential victims.

A case that defies the medical understanding of child abuse and maltreatment, which happened at a National Paediatrics' Hospital in Buenos Aires, Argentina, where inter/multidisciplinary work between public health professionals, courts and forensic experts helped to protect a child's life and her best interest will be presented.

Working with intrafamilial violence is a tough issue because professionals face incomprehensible and unreasonable situations: a victim whose perpetrator is the one who has given him/her life, or who has adopted him/her as his own son/daughter, or just somebody familiar to the child. The perpetrator is the very same person that the law or social convention says is the one responsible for the child's protection. From a psychological point of view, it's easier to admit assault or abuse from a stranger. Almost nobody is prepared to suffer violent behavior from familiar people. This is precisely the trap: how can anyone denounce child abuse or maltreatment when the perpetrator is no stranger to the victim? Would they believe the child hasn't consented? Would grown-ups believe that this is no set-up for the perpetrator? Are health professionals prepared to deal with this disturbing matter?

Medical staff has these same feelings when facing child abuse and maltreatment. At first glance, they usually have doubts about the real violence of the situation, they are afraid of reporting it to the authorities and being wrong, but they are also afraid of remaining silent and putting the child at risk. They fear they will make the situation worse if they report it and they also fear being exposed to liability in any sense.

All these doubts can be overcome if an interdisciplinary team works with this issue: medical doctors can help to provide the right diagnosis, psychiatrists can help understand family relations, social workers can provide alternatives for caring for the child, and lawyers can help decide the best way to protect the child's rights and make relations with courts a little bit easier.

This case is one of the uncommon cases in Argentina's criminal history of a female serial murderer: the probable fake sudden-death syndrome of both of "Jane's" sisters, which could only be determined retrospectively by a differential diagnosis based in medical records, but being almost certain when taking into account all the variables presented in the case. Both of "Jane's" sisters were the same age as "Jane" when they died. They had the same unspecified syndrome which, in "Jane's" case turned out to be phenobarbital poisoning triggered by her mother's criminal conduct. The victims were all women of the same age, "Jane's" mother worked as a medical receptionist and she had a psychopathic personality.

From professional experience, working together on this case helped achieve the best interests of the child, a constitutional principle owed to the Children's Rights Convention. When professionals from different disciplines and institutions focus on

the same case with their own view, expertise, and knowledge, they learn from each other and help open and enrich their mental horizons while aiming to give the best of themselves.

At this Argentine pediatric hospital, medical staff is trained in evidence collection and protection. The staff knows this is an important issue which will certainly help them stop the abuse, protect the child, and determine who is to be held responsible.

"Jane" and her brother are living with their grandmother at their home in Tucumán, Argentina.

Münchaussen by Proxy Syndrome, Intrafamilial Violence, Health Public Professionals

E47 The Role of Scientific Evidence Regarding a Child's Competency to Testify: Evidence Required to Overcome a Child's Presumptive Competency

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After attending this presentation, attendees will gain a better understanding of how trial courts view a child's competency and how the litigant carries the burden of proof to overcome the child's presumptive competency.

This presentation will impact the forensic science community by providing lawyers, scientists, and judges with insight into the significant role that reliable and relevant scientific evidence has in assessing the competency of a child witness to testify in a court proceeding whether the child is a victim or a witness.

States such as Pennsylvania have statutes governing the competency of a person to testify. For instance, according to Pennsylvania Consolidated Statutes, Rule 601, the general rule is that every person is competent to be a witness except as otherwise provided by statute or rule. Rule 601 enumerates specifically that a person is incompetent to testify if: (1) at any relevant time the person is or was incapable of perceiving his surroundings; (2) is unable to express himself or herself so as to be understood either directly or through an interpreter; (3) has an impaired memory; or, (4) does not sufficiently understand the duty to tell the truth. As further enumerated in section (b) of this Pennsylvania statute, the application of this rule is a factual question to be resolved by the court. The party challenging the competency of the testifying party bears the burden of proving grounds of incompetency by clear and convincing evidence or proving that the testimony is tainted.

In most states, there is no fixed age an individual must attain in order to be competent to testify. Age, standing alone, is not a sufficient basis to conclude the child is not competent to testify. Trial courts could and have found two-year-olds and three-year-olds competent to testify. A child is competent to testify if that child possesses the capacity to observe the events, recollect and communicate them, and has the ability to understand questions and to make intelligent answers with an understanding of the child's duty to speak the truth. The decision of competency, once raised, rests primarily with the trial judge who observes the proposed witness, notices his or her manner and apparent possession or lack of intelligence. The trial court has the discretion to ask questions of said witness to disclose the child's capacity and intelligence, as well as the child's understanding of the obligations of the oath. Trial court judges differ as to their approach in examining child witnesses. Some trial judges take the lead and ask the preliminary and foundational questions necessary for competency. The

attorneys in this approach would be permitted to ask questions after the trial judge completes his or her *voir dire* of the child. Other judges prefer permitting the attorneys to ask the preliminary and foundational questions on competency with the trial judge asking any follow-up questions. Experts suggest that regardless of who asks the questions, the type of specific questions is crucial to determining the competency of the child. Such questions should be non-leading or open-ended prompts that do not suggest the answer to the child. Examples of best practices in this area regarding questions will be provided to the attendees.

Moreover, a trial judge will consider whether the expert's interviewing techniques have tainted, led, or further influenced the responses of the child. Taint occurs when the memory of the child is considered to have been manipulated so that what the child remembers and what the child has been coaxed into thinking as a memory have been influenced by another.

To assist the trial judge, lawyers should provide testimony and reports from experts such as psychiatrists or psychologists who can testify regarding the child's ability to meet the competency requirements. The attendees in this presentation will learn the importance of hiring experts who can provide evidence based on reliable scientific methodology to assist the knowledge and understanding of the trial judge. The expert is expected to review the prior interviews and depositions of the child, examine the type of questions asked of the child, administer psychological testing such as the NEPSY-II, and review medical records and reports from other experts in the case as well as reports from agencies which serve the best interests of the child. The expert would discuss the issues of memory, suggestibility, and cognitive development and the relevance of these issues to evaluating the testimony of the child witness. The expert provides insight as to how a child's memory is created and how a child's memory can be altered. The attendees will learn about the great difficulty young children have in discriminating between related episodes in memory and how they are especially prone to confusing related pieces of information in memory. Forensic experts provide the written reports with their methodology and opinions analyzing and applying the factors which can likely influence the child's ability to provide an accurate, independent recollection of the events the child allegedly experienced.

Child Witnesses, Competency, Scientific Testing

E48 "Drew's Law:" The Prosecution of Drew Peterson

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After attending this presentation, attendees will understand the principles and consequences of new hearsay legislation, the details of the Peterson trial, and the pathology of drowning.

This presentation will impact the forensic science community by serving to illustrate the legal concepts surrounding admission of hearsay evidence as well as the investigation of deaths in bathtubs.

In 2007, following the disappearance of his fourth wife, Stacey Drew Peterson, a former suburban Chicago police officer, was charged with murder in the 2004 death of his third wife, Kathleen Savio. Savio had been found nude in a bathtub. She received an autopsy and her death ruled an accident. As a result of the disappearance, detectives were accused of botching the initial investigation by not collecting fingerprints, hair, or any other physical evidence at the Savio death scene. Savio was exhumed and the body subjected to two additional autopsies, one videotaped by a news team.

The first autopsy demonstrated foam exuding from the nose and mouth, pulmonary edema, water in the nasal sinus, and "washerwomen" changes to the hands — all typical of drowning. There was a small, 1.0 inch laceration to the back of the head. The lividity pattern and rigor were consistent with the terminal position of Savio in the tub. There were bruises on the lateral abdomen, anterior thigh, and buttocks with abrasions on the elbow, wrist, and finger. The heart was of normal size with a thickened mitral valve. The brain revealed edema but no additional injury. The toxicology analysis was negative. The death was attributed to drowning and the scalp laceration was consistent with a fall.

In 2008, the Illinois legislature created a new exception ("Drew's Law") that permitted hearsay statements into evidence if they were sufficiently reliable and were made by someone who was more likely than not murdered by the person being prosecuted.¹ The legislature used the common law doctrine of "forfeiture by wrongdoing," which allowed for the admission of some types of statements without any showing of reliability under the theory that a defendant gives up the protections of the hearsay rules and the Sixth Amendment confrontation clause by wrongfully causing the people who made statements to be unavailable as witnesses. Following an initial evidentiary trial in January 2010, Peterson went to trial in August 2012.

At trial, prosecutors attempted to construct evidence in support of homicidal injuries and proposed a variety of questions. Were the physical surroundings capable of causing anterior and posterior injuries? Was there sufficient force to cause deep anterior bruising and posterior head laceration? Was there any physical evidence of impact or transfer on the tub? Was the position of the body consistent with a fall?

All of the pathologist experts agreed that Savio died of drowning. Prosecutor's pathologist witnesses argued that a fall from a standing height in a tub would not be sufficient to cause unconsciousness and that the laceration on the head should be vertical or stellate not horizontal, which suggests wounds are from impacts with blunt objects against the head. Finally, they argued that the numerous injuries could not be sustained in a fall.

Defense pathologist witnesses maintained that the death was accidental in that all of the external bruises were unilateral, could be caused by a single fall, and corresponded to the terminal position of the body in the tub. They also argued that a person could be rendered unconscious after a simple fall without any identifiable injury to the brain. Additionally, they contended that an initial impact frequently would not produce blood splatter and would have "stunned" Savio, or possibly knocked her unconscious. There were numerous sites in and around the tub area, they argued, where the decedent could have sustained a head injury. The buttock injury is a postmortem drying artifact.

Peterson was eventually convicted and sentenced to 38 years in prison.

Some defense attorneys worry that the trial — the first in Illinois history to rely so heavily on hearsay — could fundamentally alter how prosecutors and defense attorneys handle murder cases in this state and around the country. The fervor to convict Peterson had unintended consequences. "The legal issues here are extraordinary," said Phil Turner, a Chicago defense attorney and former federal prosecutor. "If this sets a precedent, more people will get convicted because someone testifies that someone told them something." This presentation will discuss the pathological findings of the Drew Peterson trial and the future repercussions of the enabling legislation and precedent-setting prosecution.

Reference:

1. 725 ILSC 5/115-10.6 (West 2008).

Drew Peterson, Hearsay Evidence, Drowning

E49 Where Have You Been? Where Are You Now, and Who Have You Been Talking to? Historical Cell Site Analysis and Expectations of Privacy

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After attending this presentation, participants will learn: (1) how customers' cell phone billing records are being used for surveillance purposes by the government; (2) some of the technique's limitations; and, (3) the basic privacy concerns created by this new technology.

This presentation will impact the forensic science community by addressing the concerns of using the findings of this surveillance technique in court without demanding adequate safeguards of reliability, reproducibility, and error rates of its stated conclusions.

We all recognize that our concerns about the security of those we care about prompt us to check out where they are. We recognize that the same concerns prompt us to keep tabs on the locations of suspicious people and seemingly dangerous events. Parents ask their children where they will be and why, who will they be with and when. But children grow up. Eventually, they resist answering such questions. They develop a sense of autonomy and privacy, even when those asking do so out of love and concern. When our government asks where we have been and when, who we were with and to whom we were talking, it raises those same concerns about privacy and autonomy, but at a heightened level. We sense our autonomy as free citizens and privacy as individuals is being violated.

Today, the government no longer needs to ask these questions, because most can be answered by reading your cellular telephone provider's Call Detail Records (CDRs). As a rule, you are where your cell phone is, and your personal and professional associations are captured by your cell phone. It is as if a detective could follow you everywhere for years, note who you talk to, and who that person talks to, and record the date, time, and place of every conversation.

Government, from the NSA to any local police department, can now obtain CDRs to answer these questions, and their justification is to attempt to protect us, preserve order, and find out about dangerous situations. We are told that this is not surveillance, because in an Orwellian twist of language, no conversations are overheard or recorded. Yet, on the other hand, is it not surveillance if a detective could follow you everywhere for years, note who you talk to, and who that person talks to, and record the date, time, and place of every conversation?

While we may be more secure from some threats in such a world, we expose ourselves to other hazards. Historical and international precedent has shown such efforts have sometimes been used to control people and to limit their freedom. The government's massive use of such technology, absent from public scrutiny until recently, calls to mind James Madison's warning, "Since the general civilization of mankind, I believe there are more instances of the abridgment of the freedom of the people, by gradual and silent encroachments of those in power, than by violent and sudden usurpations."¹ American jurisprudence has recognized the importance of keeping our associations private. In *NAACP vs. Alabama*, Justice Harlan wrote, the "Court has recognized the vital relationship between freedom to associate and *privacy* in one's associations."² Indeed, the inviolability of *privacy* in group association may be indispensable to preserve the First

Amendment's freedom of association, particularly where a group espouses dissident beliefs.

In the criminal justice system, lawyers now use CDRs to present evidence in court supposedly showing very precisely where someone is when they make a call. They do this to achieve their persuasive goals. The evidence is made to look accurate and precise, while, in fact, it is not. In fact, a recent federal court limited such evidence for lacking scientific scrutiny.³ After attending this presentation, attendees will learn how cell phone surveillance is being used, some of its limitations, and the basis for concerns about privacy created by this new technology.

References:

1. James Madison, speech in the Virginia Convention, Richmond, Virginia, June 6, 1788, *The Papers of James Madison*, ed. Robert A. Rutland and Charles F. Hobson, vol. 11, p. 79 (1977) (emphasis added).
2. *NAACP v. Alabama*, 357 U.S. 449 (1958).
3. *United States v. Antonio Evans*, Case No. 10 CR 747-3, Northern District of Illinois – Eastern Division, Opinion and Order filed 08/29/12).

Cell Phone, Surveillance, Privacy

E50 Why Would an Innocent Person Confess? How DNA Has Proven the Unreliability of Certain Confessions and Interrogation Tactics

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After attending this presentation, attendees will have a brief overview of documented false confessions, the problems with how some interrogation tactics lead to a seemingly true confession from an innocent person, and how a false confession shapes a criminal investigation and leads to a conviction. This presentation will focus on the case of John Watkins, who confessed to a rape but who was later exonerated by DNA evidence. This presentation will educate the legal and forensic community by discussing interrogation tactics that sometimes result in false confessions which, consequently, shape the criminal investigation and conviction — often in the face of other contradictory evidence.

This presentation will impact the forensic science community by exploring how the justice system can become better equipped to weed out the true confessions from the false confessions, not only with changes in the methods law enforcement uses to elicit such confessions, but also through the use of DNA evidence.

A confession is "probably the most probative and damaging evidence that can be admitted."¹ Why would a person confess to a rape, or worse, a rape and murder, if he was innocent? And, how could this person know details about the crime if he wasn't there?

For the last two decades, DNA exonerations have shown that innocent people confess to crimes they did not commit. According to the Innocence Project's statistics, 80 of the 311 DNA exoneration cases (25.7%) involved an admission or confession from an innocent person.² In many of the DNA exoneration cases, the confessions were deemed voluntary and were sometimes corroborated by other evidence or even a co-defendant.³

The alarmingly high number of proven false confessions illustrates that interrogation techniques employed by law enforcement do not always lead to true, reliable confessions. The Reid technique is the common method of law enforcement training on how to analyze a suspect's behavior and to interrogate a suspect

in order to obtain a confession. Research has shown problems with both law enforcement and the courts being able to detect a false confession obtained through the Reid technique.³

Arizona is no stranger to false confessions. In 2003, John Watkins, a 19-year-old from Texas, moved with his parents to Gilbert, Arizona. Within the first week of their arrival, a woman had been raped in their neighborhood. The case went unsolved for ten days before John became a suspect due to discovery of his pornography stash. A police interview which began as a few questions about pornography soon turned into a four-hour interrogation on the rape case. After 68 denials of culpability, constant interruptions by police each time John tried to explain himself, being told he failed a lie detector test, that a DNA test could prove whether or not he was the rapist, that his alibi did not check out, that witnesses had identified him, and that his prints matched those left at the crime scene, John Watkins admitted he did the rape. The police and prosecutor had no doubt they had their man. The confession was the State's strongest evidence against Watkins and he was convicted.

Seven years later, DNA testing on the rape kit samples showed Watkins did not commit the crime. This meant the confession could not be true. Police interpretation of Watkins' body language was wrong. Police use of minimization tactics, promises of leniency, appeals to his moral character, and justifying the act were not tactics that ultimately extracted a confession from a criminal — but rather the lies about evidence, creating a sense of hopelessness, and accusing Watkins of lying when he denied the accusations ultimately broke Watkins down and led to a false confession.

This presentation will explore how the justice system can become better equipped to weed out the true confessions from the false confessions, not only with changes in the methods law enforcement uses to elicit such confessions, but also through the use of DNA evidence.

References:

1. *Arizona v. Fulminante*, 499 U.S. 279 (1991)
2. <http://innocenceproject.org> ("Know the cases / search the profiles")
3. Brandon L. Garrett. *Convicting the Innocent: Where Criminal Prosecutions Go Wrong*, p. 14-44. 2011.

DNA Evidence, False Confession, Wrongful Conviction

E51 Strategies for Defendant Notification in the Wake of a Major Nonconformance in a High-Volume Forensic Discipline

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After attending this presentation, attendees will understand how stakeholders can work collaboratively to provide notice to defendants in the wake of a high-volume forensic failure. The specific example used will be a major forensic nonconformance in Texas in the controlled substance discipline, where 36 counties and 4,944 cases in Texas were potentially affected by a single examiner's misconduct. Participants will also learn about stakeholder efforts to establish a statewide notice protocol for use in future similar cases.

This presentation will impact the forensic science community by addressing challenges and solutions for effectively notifying defendants about major forensic nonconformances where thousands of cases may have been impacted in a large and diverse state, as well as strategies for providing access to competent legal resources for affected defendants.

The Texas Forensic Science Commission (Commission) is charged by statute with investigating allegations of negligence and misconduct in the state's crime laboratories. The statute also requires crime laboratories in Texas to self-disclose facts to the Commission that may indicate negligence or misconduct in the laboratory. After conducting a thorough investigation, the Commission must release a public report describing the alleged negligence or misconduct and issuing recommendations for corrective action as appropriate.

In one recent example, the Commission conducted a comprehensive review and released a report describing misconduct by a forensic examiner in a controlled substance case. The nonconformance in that single case raised questions about the integrity and reliability of the examiner's work in close to 5,000 other cases. Prosecutors, defense attorneys, and members of the judiciary faced significant challenges in achieving effective defendant notification in the dozens of Texas counties affected, some of which covered large urban areas while others were located in smaller, more remote rural areas.

To address concerns regarding notification with such a large and diverse group of counties, the Commission brought affected stakeholders together to discuss the best approach. The group included the Texas District and County Attorney's Association, Texas Criminal Defense Lawyer's Association, the Office of Court Administration, the Commission on Indigent Defense, and the Innocence Project of Texas. Each stakeholder group agreed to play a role in reaching out to affected prosecutors, regional presiding judges, defense attorneys, and others affected. Letters and emails were sent to affected prosecutors, and members of the defense bar are still engaged in the ongoing process of reaching out to defendants (after the initial prosecutor contact) to provide effective notice and access to representation, especially in smaller counties.

In early 2013, the Commission and Texas Criminal Justice Integrity Unit (Integrity Unit) recognized the issue of defendant notification in high-volume cases merited a discussion among a broader group of stakeholders, with the hopes of establishing consensus for a notice protocol for future cases. In fact, it soon became evident that Texas was not alone as the Commission began receiving calls from other states facing similar high-volume notification challenges.

In July 2013, the Commission and the Integrity Unit convened a meeting of more than sixty forensic science stakeholders representing crime laboratories, prosecuting attorneys, defense attorneys, the judiciary, law enforcement, policy makers, and policy advocates. The purpose of the meeting was to discuss strategies for establishing a notice protocol for future cases, and to determine how to streamline the process and realize increased efficiencies in a state that strongly values local and decentralized control.

Recognizing that a state-funded public defense system is highly unlikely in Texas, participants identified existing state agencies and organizations that can play a greater role in the notification process. Participants also emphasized the importance of notice redundancy, and suggested many enhanced training opportunities to ensure stakeholders understood their respective roles. The group discussed special challenges faced by rural prosecutors and strategies for addressing them. The Commission and the Integrity Unit also plan to create a training video on the impact of *Brady vs. Maryland* that is specifically geared toward forensic scientists.¹ The Texas State Bar will assume a greater role in identifying and training attorneys on forensic science issues with

specific focus on how to effectively process an appellate writ in a forensic nonconformance case. Finally, participants emphasized the critical importance of educating members of the legislature and the public on these issues. These strategies will be discussed in detail to encourage creative and collaborative responses by affected parties in other states as they face similar challenges.

Reference:

1. *Brady vs. Maryland*, 373 U.S. 83 (1963).

Forensic, Nonconformance, Notification

E52 The Admissibility of Human Scent Evidence: From Bloodhounds to the Body Farm

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After attending this presentation, attendees will have a better understanding of the current case law regarding human scent evidence and the issues presented by such evidence under either a *Daubert* or *Frye* analysis.

This presentation will impact the forensic science community by giving prosecutors, defense attorneys, judges, and forensic scientists a better grasp of the legal issues presented by the proposed use of human scent identification evidence.

Dog evidence to identify the scent of a human has been offered at trial by the prosecution in a variety of cases. Some courts originally held that evidence that a tracking dog connected the defendant to some physical evidence was admissible with a proper foundation.¹ Other courts have been skeptical of dog tracking evidence as admissible evidence of guilt for a variety of reasons.² Some have felt that it was a form of hearsay while others described it as a "bloodhound myth."^{3,4} Even before *Daubert*, there was concern about the reliability of dog scent identification.⁵ Post-*Daubert* analysis of dog scent evidence has not been positive as to human scent identification by dogs.⁶

In the recent high-profile *Casey Anthony* case, the government took human scent identification to another level.⁷ It offered the testimony of a forensic anthropologist who claimed he had perfected a process of "human decomposition odor analysis." The police gathered air from the defendant's car and the anthropologist used gas chromatography to conclude that 79.2% of the gases were consistent with a decomposing human body.⁸ Florida was a *Frye* state. The judge conducted a hearing and admitted the testimony, holding that as long as the technique is generally accepted, the expert's opinion need not be. Thus, since gas chromatography and mass spectrometry are generally accepted, the expert's claimed ability to analyze the data from those instruments need not be.⁹

The judge went even further and held that it was "common sense" that "to some extent, all of us have organoleptic expertise" and that the anthropologist "based upon his background and experience could offer testimony concerning the odor he smelled emanating from the sealed container."¹⁰ He said that first responders and others are lay experts whose noses can detect the odor of a decomposing human body.

Because the jury acquitted Ms. Anthony, those opinions will never face appellate review. The case is, however, illustrative from a forensic science evidence viewpoint. The "human decomposition odor analysis" testimony would not be admissible under *Daubert* criteria. There are no standards, no testing, no error rate, and the theory is not generally accepted.

Admissibility seems less likely under *Frye*. Only requiring

that instrumentation be generally accepted for conclusions from that data to be admissible is novel at best. Further, the idea that any first responder would have such highly developed olfactory senses that they can immediately identify the smell of a decomposing human body would strain even the most ardent rejecters of the *Kumho* case.

The *Anthony* case demonstrates the outer limits of the admission of virtually any "scientific" prosecution evidence. The outcome may also demonstrate that jurors will find it overreaching and not to be, as the judge said, "common sense."¹⁰

References:

1. *United States vs. Gates*, 680 F.2d 1117 (6th Cir. 1982); *State vs. Roscoe*, 700 P.2d 1312 (Ariz. 1984); *United States vs. McNiece*, 558 F.Supp. 612 (E.D.N.Y. 1983); *People vs. Craig*, 86 Cal.App.3d 905 (1978).
2. *Brott vs. State*, 97 N.W. 593 (Neb.1903).
3. *State vs. Storm*, 238 P.2d 1161 (Mont. 1951)
4. *State vs. Storm*, *supra*: McWhorter, J.C., *The Bloodhound as Witness*, 54 Am. L. Rev.109 (1920); *People vs. Cruz*, 643 N.E.2d 636 (Ill. 1994)
5. Taslitz, Andrew E., *Does the Cold Nose Know? The Unscientific Myth of the Dog Scent Lineup*, 42 Hastings L.J. 17 (1990)
6. *Winfrey vs. State*, 323 S.W.3d 875 (Tex. Crim. App. 2010); *State vs. Dominguez*, 2011 WL 3207766 (Tex. App. 2010); *State vs. Smith*, 335 S.W.3d 706 (Tex. App. 2011).
7. *State vs. Anthony*, Ninth Judicial Circuit of Florida, Case No. 48-2008-CF-015606-O (2011), motions and orders available online at <http://www.ninthcircuit.org/news/High-Profile-Cases/Anthony/orders&motions.shtml>.
8. *Id.*, *Motion to Exclude Unreliable Evidence* (Dec. 29, 2010).
9. *State vs. Anthony*, *supra*, *Order Denying Motion to Exclude Unreliable Evidence* (May 7, 2011).
10. *Id.*, at pp.19-20.

Human Scent Evidence, Odor Analysis, Admissibility



Odontology



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F1 Evaluation of Bitemark Cases Between 2003-2013 in Turkey

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After attending this presentation, attendees will be able to utilize data presented to evaluate prevalence of bitemarks and the corresponding criminal activities in autopsies performed in Turkey.

This presentation will impact the forensic science community by showing the importance of bitemarks in forensic evaluation and in criminal investigations.

Forensic odontology takes advantage of people's unique features of jaws and teeth to determine their identities. Bitemarks, especially in the absence of witnesses in cases of sexual assault or homicide, are as important evidence of crime as are fingerprints, blood stains, and semen samples. Presence of blood and other body fluids of an attacker in a crime scene is very important. Using bitemarks to identify the presence of a second person (or persons) can be valuable evidence for defense or the prosecution team. The development of forensic odontology studies did not start until 1993 when widespread use of bitemark analysis by forensic medicine institute studies was initiated. This provided for the introduction of forensic odontology as an educational option; however, as of today, there is not a sufficient enough number of forensic odontologists in Turkey. One major reason for this shortage of expert forensic odontologists is the lack of a centralized legal forensic expert structure.

Bitemarks have been used to solve criminal cases in developed countries for a long time. Use of bitemarks as evidence is a recent phenomenon in Turkey. Evaluation of bitemarks using the newest techniques and scientific studies has contributed to the solution of criminal incidents in the world.

The Department of Forensic Odontology of Turkey's council of Forensic Medicine (ATK) is the responsible entity to identify and study bitemarks. This study evaluated 22 bitemark cases within the past 10 years in Turkey by discussing their shortcomings and comparing these result to those obtained using new techniques. There were 13 homicide cases, eight injury cases, and one bitemark case handled by the Turkish court system between years of 2003 and 2013. Four out of a total of 22 studied cases were child abuse cases, eight cases of rape, one homosexual relationship case, and the remaining were for a variety of crimes. Even though photographic analysis of bitemarks are very important in every criminal case, forensic odontologists are not present at all bitemark crime scenes. Most of the time, crime investigation teams in municipalities do not collect forensic evidence according to acceptable standards. Therefore, evidence collected at the crime scene by the investigators and especially photographs taken by this team create difficulties for forensic odontologists. One major problem with these photographs is the angle used while taking photos which create difficulty in using these pictures for

measurement and scale for analysis. The difficulties encountered in the analysis of bitemark in these cases are featured in this study.

Forensic Odontology, Bitemarks, Identification

F2 Interdisciplinary Document Toolkit for Forensic Odontological Investigation

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After attending this presentation, attendees will further appreciate the advantages of incorporating a methodical checklist approach to investigations and will understand the unique role of the forensic odontologist in the execution of the specialized expertise search warrant.

This presentation will impact the forensic science community by providing a comprehensive toolkit encompassing all the documentation necessary for a complete and methodical forensic odontological investigation, including shell forms adaptable to individual jurisdictions and situations.

Forensic odontologists are often called upon to assist law enforcement with the collection, documentation, and evaluation of physical evidence in patterned injury and bitemark cases. Individual detectives may have limited experience working a case involving a forensic odontologist and may be unsure as to what materials can be evaluated or what evidence can be collected. A checklist of records that can be useful to the forensic odontologist includes all crime scene evidence, medical reports, and more. This checklist will not only provide the odontologist with all the evidence available in a case, but will also be a ready reference for both law enforcement regarding what was provided and for the odontologist in preparing a report.

Law enforcement personnel unfamiliar with forensic odontology may also have limited experience in drafting a complete search warrant to obtain dental evidence. There exists a general search warrant, explaining the affiant's probable cause and the items to be seized, such as in the recovery of stolen property, sex crimes, homicides, and arrest warrants.¹ There are further specialized expertise "shell" warrants utilizing the specific language of the case, such as fraud (forensic accounting) and computer crimes (information technology).

However, to date there does not exist a specialized expertise shell warrant for forensic odontology, utilizing specific dental and forensic terminology and enumerating specific evidence to be collected, such as dental impressions and bite exemplars. This toolkit provides a specialized expertise shell Application and Affidavit for Forensic Dental Search Warrant, which can be adapted and individualized to comply with the requirements of different jurisdictions. This warrant takes into account the unique position of the forensic odontologist, since normally law enforcement holds the only authority to execute the warrant, collecting and documenting the evidence; in this singular situation, it is the forensic odontologist who collects, documents, and interprets the evidence.

The vast majority of forensic odontologists are also engaged in the practice of dentistry, where there are numerous patient examination forms to complete. It is understood that a complete and thorough examination is best made when each step is undertaken in the same methodical way each time, ensuring no detail is overlooked.^{2,3} This toolkit provides a forensic evaluation checklist, enumerating all evidence to be collected in a thorough investigation. This form guides the odontologist in the systematic collection of evidence and delineates and documents those items collected as a ready reference.

Sometimes records are obtained from an individual on a voluntary basis and without the compulsion of a search warrant or court order. To explain the process of evidence collection to the individual subject and to protect the forensic odontologist who collects such evidence, an Informed Consent for Examination form is supplied in this toolkit.

To define the obligations of both the forensic odontologist and the agency requesting his/her services, a Professional Services Agreement is offered in this toolkit.

Conclusion: By standardizing a systematic approach to information and evidence gathering, both law enforcement and the forensic odontologist can help ensure thoroughness, continuity, and consistency in every case. Each form is intended to be a customizable "shell" to adapt to differences in jurisdictions, situations, and personal preference.

References:

1. Herschaft, E., et al, editors. Manual of Forensic Odontology. 4th edition. Impress Printing and Graphics, 2007.
2. Senn, D., et al, editors. Forensic Dentistry. 2nd edition. CRC Press, 2010.
3. Brachlow v. State, 2005 WL 1750725, Florida 4th DCA.

Forensic Odontology, Expertise Search Warrant, Documentation

F3 Maximum Jaw Opening (mm), Prevalence, and Width of Maxillary Diastemas of Children Ages 2-18 Years

Rhea M. Haugseth, DMD, 2155 Post Oak Tritt Road, Ste 450, Marietta, GA 30062*

After attending this presentation, attendees will understand the range of maximum jaw opening of the mandible relative to the maxilla in millimeters of various age ranges with the average noted for each group. Statistical relevance and variances will also be determined. In addition, the prevalence of maxillary central incisor dental diastemas and the size measurements of the diastema were gathered and then analyzed by age and gender.

This presentation will impact the forensic science community by discussing the results which suggest that additional study may be needed to evaluate the data and its application to pattern injury analysis.

Regarding maximum jaw opening, measurements were gathered from a random group of 500 subjects over a six-month period in 2013 in a pediatric dental office. This study was undertaken to evaluate the maximum jaw openings (in mm) of children age 2-18 years old. The study was completed to determine if there exists a difference in maximum opening of the mandible relative to the maxilla in children and adolescents compared to adults. If so, can an average range of maximum opening be determined and segregated into age groups? Is there a minimum and maximum measurement of maximum jaw openings that can be determined for each yearly age of a child from age 2-18.

This information may be of use when analyzing bitemarks, possibly helping to determine inclusion or exclusion of various suspects based on age. The expectation is that there is a difference of maximum jaw openings relative to age, but not for gender.

Regarding width of maxillary diastemas, measurements were gathered from a random group of 500 subjects over a six month period in 2013 in a pediatric dental office. Data was collected as to the existence of a maxillary diastema between maxillary central incisors and its measurement for the same subject. Data for each subject's year of age and gender was also gathered, evaluated, and compared. While the existence of multiple spacings of the maxillary anterior teeth is common in the primary dentition, this study focused only on the space between maxillary central incisors as being a more distinctive and unusual finding. There is an expectation that the presence of a maxillary central incisor diastema in the primary dentition will not be different for gender or age. The presence of a maxillary central incisor diastema in the mixed and permanent dentitions may appear less frequently, in ages 6 through 18 years, due to the tooth size increase of permanent central incisors versus primary central incisors. The eruption of permanent maxillary incisors may eradicate any spacing present in the same subject in their primary dentition. This study did not attempt to monitor or note this change. All measurements were taken in one specific moment of time for each individual subject. The expectation is that there will be no statistical difference in the subjects relative to gender for the findings of a maxillary central incisor diastema in the primary, mixed, and permanent dentitions.

The results suggest that additional studies may be needed to evaluate data from more subjects and its application to pattern injury analysis.

Diastemas, Jaw Opening, Pattern Injury

F4 Just in the Lick of Time: A Case Presentation on the Importance of Salivary Secretion Swabbing Following an Acute Sexual Assault

Marnie L. Sperling, DMD, 10 Lafayette Avenue, Apt 220, Morristown, NJ 07960*

After attending this presentation, attendees will gain an understanding about the significance of swabbing salivary secretions in a timely manner following a case of acute sexual assault in regard to yielding amylase and possible DNA.

This presentation will impact the forensic science community by providing a case study and evidence-based literature regarding the DNA yield from the swabbing of salivary secretions in reports of sexual assault where the victim was either bitten or licked based on examination and/or victim report. This presentation will also strive to impact the forensic science community by broadening the understanding of the importance of detecting salivary secretions by the use of an alternate light source in addition to victim reporting in order to collect salivary amylase in a timely manner. The impact on the forensic odontology section will be to emphasize and enable a better appreciation of the outcomes in sexual assault criminal cases, which involve bitemark evidence. These cases can be better served by evidence with objective, quantitative data such as the presence of salivary amylase and DNA, rather than relying entirely on the subjective, qualitative data, such as a bitemark comparison analysis.

In instances of sexual assaults, the exchange of fluids can be seminal and/or salivary. According to the Texas Attorney General's office, many offenders have sexual dysfunction and do not ejaculate during the sexual assault. In a review of the literature,

studies have indicated that there is no ejaculation in over 50% of the sexual assault cases that are reported. Also taken into account is that many offenders use prophylaxis, have low sperm counts, or ejaculate elsewhere than on the victim's body. In cases where there is the presence of a bite mark and/or salivary secretions by either victim report or are visualized by the use of an alternate light source, it has been shown through case reviews that when properly collected in a timely manner, the yield of DNA through salivary amylase can become important evidence in cases of acute sexual assault.

This presentation will provide a case study as an example involving a sexual assault that was only oral in nature. When utilizing the American Board of Forensic Odontology's double-swab salivary secretion collection method within three hours of the assault, the evidence showed the presence of salivary amylase in two of the four collection sites which then further yielded DNA, assisting in the successful prosecution of the offender.

In conclusion, many times the successful prosecutions lie entirely on the evidence and it is imperative that forensic odontologists advance their training and are encouraged to conduct research and obtain statistics in evidence that is more objective and quantitative as opposed to subjective and qualitative.

Salivary Amylase, DNA, Sexual Assault

F5 Trafficking in Human Beings: The Role of the Dental Team in Identification

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After attending this presentation, attendees will have an understanding of the issues arising from the phenomena of Trafficking in Human Beings (THB), its correlations with dental care scenarios, and how dentists can contribute to the identification and protection of trafficked victims.

This presentation will impact the forensic science community by highlighting the importance of the interdisciplinary work between forensic professionals, law enforcement officers, social care providers, and healthcare providers regarding the identification of trafficked victims and the criminal organization behind such trafficking.

Trafficking in human beings is considered a modern form of slavery. Most oral health professionals are not aware of this phenomenon. It is the third-largest form of organized crime after drug dealing and arms sales. The first report of the European Union (EU) Commission on the victims of THB in Europe reveals that there have been 23,632 global victims in the EU from 2008 through 2010.

Italy is a destination or transit country for men, women, and children who are subjected to force, fraud, or coercion for the purpose of sexual exploitation or forced labor. The goal of governmental and Non-Governmental Agencies (NGAs), which are either directly or indirectly involved in combating trafficking in human beings, is the identification and referral of victims of trafficking and also to encourage self referrals. Identification is the most important step towards providing protection and assistance to victims of trafficking. Victims of human trafficking often have multiple physical and mental health needs which may include psychological trauma, injuries from violence, head and neck trauma, sexually transmitted infections and other gynecological problems, dental/oral problems,

and a poor nutritional condition. Healthcare and dental settings may offer opportunities for the identification of trafficked victims and oral health professionals, along with other health care providers, could play a part in recognizing, protecting, and reporting possible victims of trafficking.

Experiences in the field of community dentistry are presented here through the dental services offered to non-EU patients in Bari, Italy. Dental professionals and forensic experts can, in fact, contribute to the identification, assistance, and protection of trafficked persons, as well as offering forensic services to assist police investigations in order to identify crimes and locate the criminal organizations behind them. A check list of behavioral signs and general health indicators are proposed to oral health professional to raise awareness and contribute to the interdisciplinary approach of recognition and identification of trafficked victims.

As for domestic violence and child abuse cases, there are ethical concerns involved in the identification and protection of the trafficked persons, and a need for interdisciplinary work between law enforcement officers, social care providers, and health and forensic professionals. It is recommended that adequate training be given in forensic sciences and behavioral science, augmented by intercultural sensitivities, thus helping the dental team in the identification of these patients in order to fulfill both medical and ethical obligations.

Human Trafficking, Community Dentistry, Human Rights

F6 Case of Mistaken Identity Shatters Families

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After attending this presentation, attendees will understand the importance of scientific identification as part of the medical examiner process.

This presentation will impact the forensic science community by presenting procedures developed to prevent misidentifications and the family grief that follows.

A recent case in Phoenix, Arizona drew national attention and was the precursor for a state Senate bill that was established to help address identification mix-ups and improve steps for the positive identification of decedents. Such mix-ups are rare but are more likely to occur when officials rely on a visual identification rather than a scientific identification.

Indiana authorities visually misidentified a college student in a van crash in which five victims perished. One of the women died as a result of the trauma and was buried by her family, while a second woman who strongly resembled the decedent, survived in a coma for weeks. Upon regaining consciousness, she scrawled her name on a notepad revealing the name of the woman that was buried, shocking both families. A best-selling nonfiction book was written by the families describing this case.

In Michigan, a drunk driver struck two 14-year-old males while they were walking home from a skateboard park. The mother of one of the boys, Child A, claimed her son at the scene and rode to the hospital in the ambulance with the child who then died hours later. Child A was buried at a local cemetery after close family members viewed the body at the funeral home. The other boy, Child B, died in the emergency room and was claimed by his mother and father and was later cremated after an open casket visitation. During this service, many students from the school voiced their opinion that the boy in the casket was Child A and not Child B, but the parents denied their claims. Both children had severe head injuries and fingerprints were taken on both. One year later, the

mother of Child A came to believe she had claimed the wrong child after reading the description in the autopsy report. Child A was exhumed and a comparison of dental records proved she was correct. Fingerprint and dental record comparison demonstrated the two cases were switched. Both sets of parents had visually misidentified their children. Similar cases have been reported in New Jersey, Kentucky, and Florida.

Identifying victims who have been badly injured or killed in a car accident with multiple victims is challenging, especially with the chaos at the accident scene or at the hospital and when dealing with families desperate for information about their loved ones. Officers arriving at the scene may find the victims unconscious or trapped in the wreckage with their personal effects such as purses, wallets, clothing, and driver licenses strewn on the ground. Police and Emergency Medical Services (EMS) focus on taking care of the injured before asking questions and may have trouble distinguishing between victims who may look alike. Even family members, dazed during the aftermath and suddenness of the accident, can be uncertain.

In Phoenix, a group of five friends, one male and four females were returning from a weekend trip to Disneyland. The SUV they were riding in blew a tire and rolled over on the Interstate while traveling at 75 mph. One female died at the scene, while two females and one male were transported to the hospital. The male passenger died at the hospital the next day. The female driver was not hospitalized.

The deceased female was transported to the Maricopa County Medical Examiner's Office and admitted under the name found on one of the driver's licenses recovered from the scene. Family members of one of the hospitalized girls were informed by a nurse that their daughter had survived and was critically injured with severe head trauma. They remained at the bedside while the other family began funeral arrangements for the female awaiting the autopsy.

Upon completion of the autopsy, and at the request of the medical examiner, a dental examination was completed including a full mouth series of radiographs to verify the identity of the female who allegedly died at the scene. The medical examiner was concerned about the similarities between the photographs of the two driver's licenses recovered at the scene and wanted confirmation by scientific means.

Comparison of the antemortem dental radiographs provided by the allegedly deceased female's family and the postmortem dental radiographs obtained by the forensic odontologist revealed numerous inconsistencies. Antemortem dental radiographs of the second victim (purportedly in the hospital) were compared to the postmortem dental radiographs and scientific identification was confirmed. All parties were immediately notified, especially the two families of the females involved in the accident.

This case of mistaken identity led to a Senate bill in the State of Arizona called "Abby's Law" which allows the family, on request, to view a decedent within 48 hours in order to establish positive identification. This law does not preclude misidentifications, but it does provide a reasonable timeframe within which a suspected mix-up can be addressed.

Lessons learned from this case include: (1) visual identifications may be challenging and unreliable; (2) protocols should be established to include scientific identification in predetermined scenarios; and, (3) emotion surrounding these types of cases can lead to legislative oversight which has more far-reaching effects on the practices of the medical examiner.

Mistaken Identity, Scientific Identification, State Senate Law

F7 I Dent: Leveraging Simple Technology to Facilitate Dental Identification to Access the Expert Wherever They May Be Hiding

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After attending this presentation attendees will: (1) understand the concept of distance identification and when it should and should not be used; (2) be able to use commercially-available hardware and software materials to carry this out in their own autopsy suite; (3) supervise associate forensic odontologists during their training; and, (4) understand the role of the pathology assistant in the process of facilitating dental identification.

This presentation will impact the forensic science community by providing a simple guide to methods that allow coordinated efforts between advanced practice pathology assistants in a secure setting and the forensic odontologist in instances where direction of the oral autopsy, guidance for image selection, and interim identification by dental means is required.

The function of a medical-legal autopsy is to determine the means, manner, and cause of death and, importantly, the identity of the deceased. It is well documented that dental identification is simple, cost-effective, and efficient in the process of body identification. Performing critical procedures at a distance from the patient is well-established in medicine and includes tasks as variable as electrocardiogram interpretation, diagnostic image interpretation (telerradiology), and even robotic surgery. This study has extended this concept to the process of dental identification, specifically regarding distance dental identification that may be undertaken in cases of dental identification when the forensic odontologist is at a distance from the autopsy suite. It is dependent on establishing a secure and nimble means of communication and simple file transfer.

Initial contact is made with the forensic odontologist of a pending case, using smartphone technology. Included in this communication are the case number and putative identification. A specific time is designated for further secure communication using password-protected technology. Once antemortem records are received, the pathology autopsy assistants, with advanced training in dental radiography and anatomy, submit the antemortem records via images attached to texts, e-mail attachments, or directly using commercially available software. The forensic odontologist verifies the case number and, if necessary, directs the clinical and radiographic examination and data collection. The results of the postmortem examination are sent similarly to the antemortem data. Once received, the forensic dentist uses a commercially available smartphone projector to: magnify and evaluate both the antemortem and postmortem data; antemortem and postmortem clinical and radiographic data for points of concordance; and, to determine if the case is identified. Subsequently, a report is prepared and sent. Alternately, the body can be held until the forensic odontologist can personally attend or a back-up forensic odontologist can be called if the case is a thorny one or one that requires multiple complex imaging. Once images are digitally captured and transmitted, a case can be sent to a trainee odontologist, who may simultaneously prepare a draft report for review by the senior odontologist.

The most basic material set for this to be carried out is two smartphones, but may include computer tablets, portable projectors, or laptop computers. Issues to be aware of include transmission privacy, security, transmission of data from both in the mortuary and to geographic locations where cell phone coverage is problematic. Additionally, for identifications undertaken outside

Canada, it is necessary to work in cooperation with Embassy staff to ensure contact with the supervising forensic pathologist or investigating coroner.

The system is simple, inexpensive, robust and has been used in geographic locations in Canada, United States, South America, and at sea in the Pacific. It is not intended to replace "live" identifications, but to act as an adjunct, a training tool, and as means of undertaking peer review and precludes the necessity of removing jaws from remains to transport to the forensic odontologist.

Dental, Identification, Technology

F8 Dental Identification in a Maxillary Edentulous Individual Missing for Seven Years

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After attending this presentation, attendees will understand how identification of an unidentified decedent is often a collaborative effort between the forensic odontologist, forensic anthropologist, forensic pathologist, and law enforcement.

This presentation will impact the forensic science community by showing how a positive identification by the forensic odontologist and anthropologist can be made utilizing an edentulous skull.

On February 15, 2013, a man hunting for deer antlers found an edentulous skull with no mandible in a wooded area of Brownstown Township, Michigan. The only missing person in this community disappeared in 2007 under suspicious circumstances. At the time of her going missing, her home was set on fire and her luggage was found in her vehicle, which was also set on fire. Law enforcement has long suspected her husband in her disappearance, which they believed was a result of foul play. After the disappearance, law enforcement opened an investigation into his first wife's death, initially believed to be an accidental fall. The husband is currently in prison serving time for sexually assaulting a mentally disabled woman and for the 1990 killing of his first wife, for which he had been convicted of second-degree murder. He denies any involvement in the disappearance of his second wife.

At the time of the initial investigation by law enforcement in 2007, the Brownstown Lt. Detective Robert Grant recovered a maxillary complete denture at the fire-ravaged home. The detective went to the missing individual's dentist and received a written statement that this denture was the missing woman's. The victim was recently evaluated in his office for a crack in the denture. The detective also received and stored all missing individuals' dental records since 2007.

Upon examination of the skull by the forensic anthropologist, a biological profile was developed. This skull was determined to be that of a Black female, age 35- to 50-years-old. A maxillary torus and right temporomandibular joint abnormality was documented.

Upon review of the dental records and examination of the skull by the forensic odontologist, a positive identification was made. This was based on the shape of the large and atypical maxillary torus, having been reduced and flattened by an oral and maxillofacial surgeon before the fabrication of the denture. This shape was reflected in the previously identified complete denture. The antemortem dental radiographs from the dentist also showed a large radiolucency at the apex of tooth #6, the maxillary right

cuspid. This was also seen in the skull. Finally, the antemortem panorex showed some bony changes on the right mandibular condyle which corresponded to the right temporomandibular joint abnormality seen in the skull.

A week later, law enforcement and the forensic anthropologist returned to the area to conduct another search. At that time, the mandible and additional skeletal remains were recovered. The existing mandibular teeth were then compared to the antemortem dental records and once again the identification was confirmed. The right condyle had the same abnormality seen on the skull and the antemortem panorex.

This case reinforces the collaborative effort of law enforcement, forensic odontology, forensic pathology, and forensic anthropology in the identification process.

Dental Identification, Edentulous, Maxillary Torus Palatinus

F9 Next of Kin Correspondence: Additional Dental Evidence Aiding in the Identification of Korean War Remains

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After attending this presentation, attendees will appreciate the utilization of additional evidence to substantiate the identification of Korean War remains.

This presentation will impact the forensic science community by reminding all scientists that all information gained, no matter how minimal or when obtained, may in the future help to substantiate an identification.

The mission of the Joint POW/MIA Command (JPAC) is to conduct global search, recovery, and laboratory operations to identify unaccounted-for Americans from past conflicts in order to support the Department of Defense's personnel accounting efforts. Of the approximately 1,900 sets of remains identified by the JPAC, 224 are from the Korean War. In recent years, the proportion of identifications from the Korean War has increased and comprised 42% of all JPAC identifications from the last three years.¹

Following the end of the Korean War, Operation Glory (July-November 1954), an agreement between the United Nations and the Communist Forces (People's Republic of China and the Democratic People's Republic of Korea (North Korea)) was conducted and led to the exchange of war remains. The United Nations (UN) transferred all suspected American remains to the U.S. Army's Central Identification Unit (CIU) in Kokura, Japan, for analysis. At the CIU, the remains were examined by Graves Registration identification specialists and anthropologists. All remains declared by the CIU and approved by a military review board as unidentifiable were transferred to the National Memorial Cemetery of the Pacific (The Punchbowl) in Hawaii and interred. Over 400 sets of unknown Korean War remains were buried at the Punchbowl.

During the Korean War, the Office of the Quartermaster General's (OQMG) Graves Registration (GRREG) was responsible for the recovery, processing, identifying, and repatriation of remains. The strong commitment of the Graves Registration Service (GRS) to identify remains is reflected by their persistence to obtain additional information, specifically requests for additional dental and medical records/information from military health facilities and next of kin. If a response was not received in a timely manner, a follow-up letter was sent.

The OQMG written request resulted in a range of responses from family members to include medical/dental histories, civilian

health records/radiographs, contact information for civilian health care providers, photographs, and general descriptions of their son. While the information obtained from family member replies were not all substantive, all additional evidence was considered while writing the forensic odontology reports. It is also important to note that the assemblage of dental/medical records during this time period was not an easy task, as health care providers were not required to keep their records for an extended period of time and duplication of records would have to be performed manually.

This presentation will provide examples of additional dental evidence provided by family members and utilization of this information to substantiate forensic odontology reports.

Reference:

1. Ah Sam, FY Identifications as of 2013 02 14

Joint POW/MIA Command (JPAC), Odontology, Additional Dental Evidence

F10 Single Tooth Identification: Three Case Studies

William E. Silver, DDS, 10 Edgewater Drive, #5G, Coral Gables, FL 33133*

After attending this presentation, attendees will understand the value of a single tooth in the process of dental identification as well as in the process of exclusion during dental identification.

This presentation will impact the forensic science community by a direct effect upon those who participate in the dental identification process. Persons who are acutely interested in the identification of unknown missing persons also may be members of the law enforcement community or relatives of a missing person.

Dental identification is usually based upon multiple loci of restorations present in the teeth of the dental arch. It is accompanied by radiographic and photographic records and a comparison of the antemortem and postmortem records of a similar nature. Often these antemortem records are absent, illegible, or ignored in the process of identification. Even the postmortem records may not be created in a proper manner and result in the same inadequacy. The process as well as the material are important, but on occasion the record comes down to only one tooth. In addition, there may be an inadequate application of basic principles which may confound the identification.

Three cases are presented which demonstrate various degrees of difficulty when presented with different situations when only a single tooth is involved. In the first case, a parent unknowingly gave false information regarding the condition of a single anterior tooth which led to the mistaken identity of a person who was not the victim of an automobile accident. Subsequent microscopic examination of that single tooth produced evidence leading to the correct identity of the victim, who was in fact her son.

In the second case, which was involved in the Valujet™ airplane disaster in Miami, there was a fixed population who were aboard the plane. There were a limited number of victims and a fixed number of dental specimens available for identification. In this instance, there was a single tooth which could only be matched with one antemortem record. No other victim, whose records were available, demonstrated a restoration of that size and shape in that same tooth.

In the last case, there was an unfortunate set of circumstances leading to multiple omissions and errors which eventually developed into a single tooth identification. Neglect of cleansing of teeth and replacement of scattered teeth into their proper places contributed to the confusion. In addition, improper placement of radiographic sensors, cone cutting, and neglect to consult with the medical examiner for additional postmortem

evidence prevented the identification by traditional forensic odontological methods. Finally, neglecting all other means, the identification was accomplished by the close examination of the antemortem and postmortem records of the pattern of one restoration contained within one single tooth.

It is incumbent upon the forensic odontologist, in the absence of ideal evidence, to pursue the evidence contained even within one single tooth in order to make a positive identification.

Tooth, Odontology, Identification

F11 A Case Report of Three Beheadings in Puerto Rico

Xiomara N. Rivera, DMD, Urb University Gardens, 251 Fordham, San Juan, PR 00927*

After attending this presentation, attendees will have learned about three beheading cases on the island of Puerto Rico, where identification of the head through dental means was necessary.

This presentation will impact the forensic science community by presenting three different cases of beheadings with three different motives for these crimes (a theft, a drug-related crime, and a hate crime), thus presenting different crime scenarios where dental identifications are needed.

Beheading is defined as: "separating the head from the body." It can occur unintentionally (for example, in a traffic accident) or it can be done intentionally as means of murder or execution. Intentional beheading is known to have been commonly used in the past since it was a simple method of execution as swords were easily available. Considered a noble death in early England, it might have been preferred by prisoners to other forms of execution.

If a single blow is sufficient to completely decapitate the person, it may be considered a "humane" method of execution of a prisoner. However, for it to be so, since the cervical muscles and vertebrae are strong, a skilled headsman is required, if an unsharpened instrument is used or a person without skill commits the act, several strikes may be needed to decapitate. Thus causing the victim prolonged suffering. Some of the most famous people to be beheaded in history were John the Baptist, Anne Boleyn, Charles I, and Mary, Queen of Scots.

In our society, cutting a person's head off is considered barbaric. Since few things make a greater impression on the public than seeing a severed head, decapitation's shock value has been used to strike fear in enemies and/or ensure obedience to whoever commits or orders this act. Such is the case of a 17-year-old male who was found decapitated in a bush-filled area. Identification of the head was achieved through dental photographs and dental record comparison. The body was identified by DNA analysis.

Another incident was the crime of a 19-year-old transsexual who presented a partially burned and dismembered body, that had been beheaded. Identification of the head was achieved through dental record comparison as well.

In the last case to be presented, the victim of a theft was stabbed multiple times and beheaded in his home by his nephews. The head was placed in a different room in the house from the body and dental records were used to make the identification of the head.

These cases will be presented to familiarize the forensic odontology community with uncommon cases that require the identification of only a human head through dental means.

Beheading, Dental Identification, Puerto Rico

F12 The Use of Digital Gamma Correction Curves in Bone Trabeculation Mapping for Victim Identification

Henry J. Dondero, DDS, 2 Emerald Drive, Glen Cove, NY 11542*

The goal of this presentation is to deal with the use of digital photo editing software as an aid in accentuating boney landmarks. This will aid the forensic odontologist in being able to utilize all devices and methods available in the quest for victim identification.

This presentation will impact the forensic science community by encouraging forensic odontologists to be aware of the various investigative modalities available.

The forensic odontologist may not be able to identify every victim he or she encounters due to a multitude of reasons. One situation arises when a victim presents with few or no restored teeth. It is particularly challenging when a victim is edentulous. To receive a few or outdated antemortem radiographs further compounds a difficult situation. The following case is an example of an identification that was made based on a single radiograph that was several years old. What is particularly unique is that the boney identifiers were highlighted through the use of digital photo gamma correction editing software.

This case involved partially decomposed remains found in a wooded area near an irrigation recharge basin. The body was that of a 25 to 30-year-old male. The head and neck were badly decomposed with marked presence of insect larvae. The police had ruled this case an execution-style homicide associated with an illicit drug transaction. Initial dental examination revealed a nearly complete dentition with relatively few restorations. Radiographs were taken and clinical charting was performed. The victim had several items of identification on his person and an attempt was made to locate a family dentist to verify an identification but to no avail. A check with the victim's insurance company revealed a dental visit comprised of one radiograph and an extraction performed approximately three years earlier. The single radiograph was sent to the medical examiner's office and compared to the victim's postmortem records.

The radiograph received as the only antemortem record consisted of a periapical projection of the lower right first molar area. Teeth numbers 27, 28, 30, and 31 were present and numbers 29 and 32 were missing. All teeth were virgin except number 30 which showed an extensive carious lesion. Initial comparison with the postmortem radiograph showed great similarities, except that tooth number 30 was now missing. Because no other records were available, both images were saved as TIFF files for further comparison pending digital enhancements. The files were then opened in a digital photo editing software application and adjustments were made to the brightness, contrast, and sharpness of the image as well as enlarging certain areas of interest. Further enhancements were achieved by the use of alteration of the gamma correction curve. This last procedure elicited a striking detailed image of the bone trabeculation anatomy, especially the interradiolar area of tooth number 30. This postmortem area image, when compared to the antemortem image area, showed an exact outline of the trabeculation anatomy sufficient to ascertain identification.

The resultant enhanced images afforded the opportunity to more accurately compare antemortem and postmortem radiographs sufficient to establish identity. The use of these techniques allows the forensic odontologist the opportunity to view, compare, and, in many cases, certify identification which would otherwise remain covert. Areas of increased calcification such as retained root fragments, hyperostotic borders, and bone trabeculation patterns can be compared. Many radiolucent areas

containing unique morphology are also greatly enhanced.

Radiograph, Enhancement, Gamma Curve

F13 True Crime Stories: Odontology and Murder

Allan A. Raden, DMD, 4 Monroe Avenue, Box 863, Glassboro, NJ 08028*

After attending this presentation, attendees will better understand some of the principles involved in identification of non-viewable remains.

This presentation will impact the forensic science community by offering insight into the methodology and sequencing of investigations to successfully resolve a dramatic crime.

This presentation discusses the role of the odontologist in the investigation of a homicide. Frequently, an odontologist is called upon to make an identification of remains that may be part of a murder investigation. After a medical examiner's ruling of homicide, it is critical that an accurate and provable identification is made. The first thing a good defense attorney would do for his accused client would be to question the identity of the deceased person. The odontologist is employed to demonstrate the identity when other methods such as DNA or fingerprints are not possible or impractical.

Two cases will be presented that demonstrate the need for accurate and timely identification as part of the investigative process. Background information on the victims as well as outcome will be described. The first case involves a middle-aged woman who had an unstable relationship with her boyfriend. A neighbor witnessed an argument between the two of them which resulted in a kidnapping. The boyfriend also kidnapped a friend who tried to intervene on her behalf. He later let the friend escape, but drove across state lines and eventually beat and murdered his former girlfriend. Although charged with her murder, the prosecutors had no body as the trial date approached. A deal was struck which revealed the location of her remains. While controversial, this deal-making helped expedite the resolution of this case. The second case involves a murder, but at the time of this writing, nobody has been charged. These are relatively high-profile cases that were reported in the media for several weeks. It was through the tireless efforts of investigators that eventually led to the location of one victim. An unusual radiographic method was employed in this case, and it was fortunate that the dental evidence, though scant, was very strong. A confounding factor will be discussed.

It is hoped that this presentation will impact the forensic community by highlighting methods used to make a positive identification and a prosecutorial method that expedited resolution. When evidence is minimal, every effort must be used to make the best of it.

Homicide, Police Investigation, Radiography

F14 Dental Identification Utilizing Digital Dental Laboratory Evidence

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After attending this presentation, attendees involved in victim identification will gain better insight into identification modalities utilizing digital dental records stored in a dental laboratory

database. The introduction of new technologies has increased the potential to gather significant identifying antemortem evidence.

This presentation will impact the forensic science community by introducing an additional source of dental information that may possibly be used in a dental identification of recovered human remains. Awareness of this digital evidence may possibly provide critical information that could establish a positive identification in a previously undetermined case.

On July 13, 2011, Pennsylvania State Police (PSP) discovered the dismembered and burned remains of what they believed to be two humans. The victims were tentatively identified as husband and wife, residents of the estate where they were found. Upon further investigation, the son of the presumed male victim was arrested in connection with the deaths.

During their scene investigation, bones were discovered in and around a pond on the property, approximately 250 yards from the house. Two burn barrels were also located about 200 yards from the house. A forensic anthropology team from Mercyhurst College in Erie, Pennsylvania was called in to perform a forensic archeological survey and recovery from the scene.

Among the specimens recovered by the anthropologists was a human skull with the maxilla, a separate mandible, and a pelvis consistent with a female and one consistent with a male. Along with various other bones, incinerated fractured teeth and roots were recovered. Also, what were believed to be two fractured ceramic crowns were discovered in the burn pile.

The initial recovered skeletal and odontogenic materials that might establish a dental identification were presented to the forensic dental consultant by the PSP. The odontologist performed a complete forensic dental exam which included photography and radiography. The fragmented skeletal remains were compared to the antemortem (AM) records, narrative and radiographic, of the presumed female victim. The quantity and quality of the recovered dental evidence were sufficient to establish a positive identification.

As the scene recovery continued, further dental evidence was recovered. These were also presented to the odontologist. The quality and quantity of the postmortem materials were incinerated and insufficient to establish a positive identification. The evidence allowed for a possible, non-exclusionary identification. Antemortem records were sufficient to establish identification with adequate postmortem evidence. Among the recovered remains were two laboratory-fabricated ceramic crowns. Although similar in shape to crowns of the suspected male victim, a positive identification was not immediately possible. The postmortem evidence was insufficient.

In July 2012, the PSP presented the crowns to the State University of New York at Buffalo School of Dental Medicine based Forensic Dental Research Laboratory in an attempt to establish an identification from a dental material standpoint. The ceramic crowns were analyzed but the material composition was not sufficiently unique to establish a positive identification. Without a positive identification of the male victim, the defense submitted a motion to dismiss charges against the suspect.

A break came in the case when, through further questioning of the male suspect's dentist, it was determined that digital laboratory records existed. The crowns were fabricated through a Computer-Aided Design/Computer-Aided Manufacturing (CAD-CAM) system whereby digital records could refabricate exact replicated dental dies of the suspect's prepared teeth prior to cementation of the crowns. Recovery of this digital information and an analysis of the adaptation of the crown fragments to the dental dies established a positive relationship to the male suspect and the recovered dental evidence. This information, along with recovered male skeletal remains and other non-exclusionary dental evidence, established a positive identification of the male suspect. This identification allowed the district attorney to proceed with the charge of double homicide against the suspect.

Dental Identification, CAD-CAM, Homicide

F15 The Lac-Mégantic Disaster

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After attending this presentation, attendees will acquire information regarding the fatal train derailment in Lac-Mégantic, Quebec, in 2013 and the complementary roles that DNA and dental identification played in the disaster.

This presentation will impact the forensic science community by providing understanding of the complementary roles that DNA and dental identification play in mass disaster management.

The unattended 72-car freight train operated by the United States-based Montreal, Maine, and Atlantic Railway (MMA) carrying over two million U.S. gallons of crude oil ran away, derailed, and caught fire with multiple explosions in the center of the town, a scenario worthy of a Stephen King novel. As of this writing, 42 people are confirmed dead and five are still missing and presumed dead. There were over 30 buildings destroyed in the town's core.

Participants attending this presentation will be informed concerning the problems and the solutions involved with the fire, explosions, the effects of heat at 3,000°C for 36 hours, soil contamination, collapsed buildings, burned victims, and the logistics of organizing such a scene.

It was initially thought there would be no victims to identify because of the extreme heat and longevity of the fire. The first two bodies were recovered the following day on the periphery of the incident as the center of fire continued to burn for another 24 hours.

The key to resolving these multiple-tiered scenarios is organization and determining who does what. Controls involve knowledgeable competent personnel in every phase of the operation.

The forensic identification team on site involved two pathologists, an anthropologist, and an autopsy technician. Each case was bundled, packaged, numbered, identified, and triaged by soft tissue, bone, and dental if the body was fragmented. Pathologists, dentists, and DNA experts further analyzed the remains at the central forensic laboratory in Montreal, 150 miles away. Each bundle was photographed, examined and underwent fluoroscopy and radiography. A pathologist, a dentist, and autopsy technician performed fluoroscopy of each bundle. The latter was particularly useful in suspected commingled remains. Both hard and

soft tissue were provided for DNA analysis.

Most dental cases were severely charred. Most jaws were fragmented and individual crowns and roots had to be identified separately as they were unattached. The roots of multi-rooted teeth were frequently separated. Dental trabecular bone pattern morphometric analysis and comparison was particularly useful in identifying fragmented remains.^{1,2}

DNA and dental identification played complimentary roles. The coroner demanded at least two types of identification before declaring a death official, for example, DNA and dental identification. In addition, a peer within each discipline independently validated the results. The reasoning for such a cautious approach was twofold; correct identification of the body or fragment thereof, as well as an overall picture of where each body/fragment was found. Explosions, building collapse, and comingling prompted this unusual approach with very interesting results.

References:

1. Desranleau S, Dorion RBJ. The trabecular bone in Identification. Proceedings of the American Academy of Forensic Sciences; 2011, Chicago, IL.
2. Desranleau S, Dorion RBJ. The trabecular bone in identification - Part 2. Proceedings of the American Academy of Forensic Sciences; 2012, Atlanta, GA.

Mass Disaster Management, Positive Identification, Trabecular Bone Patterns

F16 An Identification Case Where Social Media Provided Evidence for Dental Comparison

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After attending this presentation, attendees will understand that when developing an opinion for submission to a medical examiner (ME)/coroner, the forensic odontologist will compare postmortem data to identified antemortem records. When the antemortem record is limited or absent, the odontologist has the best understanding of what can be valuable material to develop an antemortem dental profile. The odontologist can become part of the investigative team offering aid and support to law enforcement and the ME/coroner.

This presentation will impact the forensic science community by showing how an odontologist can assist in the development of a missing person profile before the discovery of remains and the comparison for identification. Collected progress notes, models, radiographs, photographs, odontograms, whitening trays, retainers, and insurance Explanation of Benefits (EOBs) are some items that provide antemortem dental information. Analysis of this material is done to develop, as completely as possible, an antemortem profile of the missing person. Scientific methodology can only be enhanced with increased availability of data. Sources of data may be family, friends of the missing person, health care providers, law enforcement, the ME/coroner, etc. Social media has become a vehicle for networking and a public database. This public display of names, places, dates, and images carries inherent privacy risks but, in this identification case, researching the family's use of social media provided unintended benefits toward identification of their lost family member.

Synopsis: A missing person was reported. Limited antemortem reference material was available from health care providers to develop an antemortem dental profile for inclusion in a National Crime Information Center (NCIC) database for potential dental comparison. An online search of sites which included the missing person's name, provided several sites of news sources and a notation of a Facebook® posting. Linking

to the Facebook® site, a posted string by family included several photos of the missing person. The photos were posted as memorial images without consideration that the images could have forensic value. One image posted long before the individual was reported as missing was distinctive from the other images by its dental detail. Law enforcement was notified. When remains were eventually found, digital images of the remains (postmortem digital photographs of the dentition) were captured in the field and digitally forwarded for comparison to the photographic image posted on Facebook® and the available antemortem radiographs. The single antemortem photograph eventually became a valuable comparison element contributing to a written report opining a positive dental identification. This scientific method of identification enhances and documents the confidence level of identification over less-scientific means of identification such as personal effects and association.

Conclusion: The public's participation and utilization of social media may provide valuable forensic data that the lay person does not appreciate as having scientific value. Scientific opinions are better formed when every source of information is explored and that information is then considered.

Forensic Odontology, Identification, Forensic Anthropology

F17 The Emergence of a Disaster Victim Identification (DVI) Team in Hungary

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After attending this presentation, attendees will gain a clearer understanding of how a Disaster Victim Identification (DVI) team was established in Hungary.

This presentation will impact the forensic science community by raising awareness that DVI teams need to be formed and well trained for the welfare of society. The establishment of a DVI team in Hungary may serve as an example of how challenging this undertaking could be, and that through perseverance and commitment such a goal can become reality.

Since 1989, as the Communist era came to an end in Hungary, mafia gangs and political groups emerged mainly in Budapest, the capital city, where violence was virtually unknown and non-existent. Among many blasts and car bombs, the terror attack on Soviet Jewish Immigrants in 1991 and the assassination of four people in downtown Budapest in 1998 were most significant. In the years to follow, traffic accidents became the main producer of disaster victims.

In 2008, the first attempt to establish a DVI team was initiated by the Semmelweis University's Forensic Department, addressing the State Secretary of the Ministry of Justice who approved the idea. However, bureaucracy, lack of interest and funds, as well as other priorities, prevented this plan from actually coming to fruition. Thus, the Medical University's Forensic Department took it upon itself to create a Forensic Expert Team as part of a DVI group which included a forensic pathologists, dentist, anthropologist, and DNA specialist. This independent initiative finally encouraged the officials of the Ministry of Justice and Interior in 2010 to appoint a DVI unit under the umbrella of the Central Crime Scene Investigation Department within the Hungarian National Police Force.

In 2011, a bus crash in Egypt which killed 11 Hungarian tourists intensified the urgency of this matter. As a result, the department in charge developed the infrastructure of DVI in Hungary, ensuring a nationwide coverage including the forensic departments of all four medical universities as well as the local forensic police expert teams.

Presently, concrete plans are underway to create a

common vision among those involved in DVI through workshops and training. International cooperation, especially within Europe, is necessary and encouraged to ensure the highest of professional standards in this field of forensic science.

In facing common challenges and service to humanity, the concepts of accompaniment and assistance in developing capacities are vital in chartering an individual path for progress and growth. Great accomplishments in public service are possible through unity of vision and action and mutual and ongoing understanding, collaboration, and support.

DVI, Hungary, CSI

F18 Postmortem Dental Examination: 3D Modeling of Dental Arches

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After attending this presentation, attendees will understand the interest in the postmortem dental examination carried out by optical impression, especially as it most often allows one to avoid the sampling of the mandible, since it presents a 3D color picture of the dental arches and backs up a large quantity of data.

This presentation will impact the forensic science community by making the experts aware of the fact that, thanks to this new protocol and the cutting edge technologies used, most postmortem (PM) dental examinations may be carried out in good conditions while complying with the forensics' ethics and the international directives. This new protocol tested in the Forensic Science Institute of the French Gendarmerie is currently used in some cases.

The objective of this study is to finalize an examination protocol in forensic odontology that corresponds with the needs and ethics of forensic odontologists while complying with the international and national directives and especially those of the International Criminal Police Organization (INTERPOL) concerning the sampling of mandibular and maxillary pieces. In the guide dealing with the identification of the victims of a disaster (INTERPOL 2009), the postmortem data collection describes in chapter 6 the characteristics of the PM dental examination. It determines the conditions of the removal of the maxilla and the mandibles. "The extraction of the jaw(s) should not be considered unless under exceptional circumstances. The odontologist must present the arguments justifying this procedure to the person in charge of the supervision of the dental examinations on the PM site beforehand..."

The virtual autopsy exists today in forensic pathology and the equipment compatible with this kind of examination starting to be used in autopsy rooms. The forensic odontology must follow this evolution and it is up to the forensic odontologists to propose the new protocols which will be approved in the next few years. They must also test the most reliable, most maneuverable, and easiest equipment to use. New technologies are there to help solve the problems of low-quality mandibular and maxillary samples during postmortem examination.

This research tested the optical impression's capacities to carry out PM examinations without resorting to dissection. A protocol was initiated in order to avoid sampling of anatomical pieces while providing as much information necessary for identification.

Materials and Method: The use of a laptop is necessary and sufficient to gather real time 3D image analysis, reconstruction and data storage software programs. An optical camera linked to this laptop by a USB port provides for the taking of impressions.

The methodology of the study differentiates several programs. First, a round of tests of impressions on dry skulls has been performed by several forensic odontologists who had received only a basic introduction to practice optical impression taking. A second program has allowed the same experts to carry out tests of impressions on living individuals lying on a fake autopsy table. These tests were intended to check the handling ability of the equipment, the accessibility to the oral cavity, and the ergonomics. Finally, a third program was initiated to validate the intervention on deceased bodies and determine the protocol.

Conclusion: The access to the dental CAD/CAM in forensic odontology gives another dimension to postmortem data collection.

The data can be exported with speed and facility both from an autopsy room to a laboratory nearby and to a processing center on the other side of the world.

If it seems possible to initially decrease by the number of samplings by 50%, the results should quickly reach a reduction of 80% in the years to come.

This work complies with forensics' ethics and the needs of experts as much as the virtual autopsy does, but it can only progress with the consent of the scientific community.

Dental CAD/CAM, Postmortem Examination, Ethics

F19 The Non-Radiographic Dental Identification of Christopher Dorner

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WITHDRAWN

Dental Identification, Charred Remains, Positive Identification

F20 Physical and Radiological Appearance of Human Teeth Exposed to High Temperatures

Lee P. Coppess, DDS, 7061 E Hayden Haven, Hayden, ID 83835*

After attending this presentation, attendees will learn of the effects of high temperatures on the appearance of human teeth at various lengths of exposure time.

This presentation will impact the forensic science community, especially those relatively new to the odontology field, by visually demonstrating varying degrees of thermal destruction of human teeth when subjected to a constant temperature (1,000°F) for varying lengths of time.

Burned human remains may be recovered from a wide variety of incidents, many of which include structural fires and vehicle fires. Likewise, these fires may include an extremely wide range of temperatures. The soft tissues of the oral cavity provide a certain degree of insulation to the teeth until they are consumed by the immolation process.¹ Certainly, due to the wide range of temperatures and exposure times that burn victims are subject to, there can be no constant values for the appearance of the teeth. In gathering the information for this presentation, human teeth were subjected to a temperature of 1000°F for ten minute-increments exposure time up to 50 minutes. Two separate sets of teeth were treated: the first set was molars, similar in size and shapes, the second set were premolars, also similar in size and shape. A dental laboratory burn-out oven was used for the heat source. A

set of two photographs and radiographs were made of each tooth, one prior to and one following their exposure to heat.

Results: There was very little difference in physical appearance of the teeth between the ones heated at ten minutes and the ones heated for 50 minutes, although as the length of time increased, there seemed to be an increase in the brittleness of the enamel of the teeth. In all cases, the enamel became extremely brittle as is documented by the photographs. Radiographically, again there was little if any difference in appearance between the exposure times, other than for the breakdown of the enamel portion of the teeth.

Conclusions: The physical and radiological appearance of teeth observed or recovered from a burn victim would likely not allow an investigator to offer an opinion as to the intensity of the heat or the duration of time that the victim was exposed to the heat source.

Reference:

1. McGivney, James, DMD. The Identification of Burned Human Remains. Proceedings AAFS, volume 18, page 266.

Teeth, Exposed, High Temperatures

F21 John Wayne Gacy — Still in the News

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After attending this presentation, attendees will review a historic case, including all ID efforts, court preparation and presentation, a current media review of unsubstantiated attorney accusations, and irresponsible journalism, in spite of current ID confirmation by twelve forensic experts, most of whom are American Academy of Forensic Sciences (AAFS) members and six of whom are board certified.

This presentation will impact the forensic science community by outlining for odontologists of all levels of expertise, the largest U.S. mass murder attributed to one individual, but primarily will emphasize current accusations which can be detrimental to one's reputation and professional success.

John Wayne Gacy was arrested in December 1978 for the murder of 33 men and boys, most of whom were buried in his crawl space. Initially 24 of the victims were identified primarily by dental means. John Wayne Gacy was tried in February of 1980 for the murder of eight of the 33 victims, was convicted, and was executed in 1994. One more victim was identified in May of 1986. In an effort to identify the remaining eight unidentified victims, they were exhumed in 2011. The exhumation effort would yield one additional identification through DNA, confirmed with dental records not previously available. At the same time, a woman whose son was identified by four forensic odontologists on the original ID team and a consulting radiologist, insisted the identification (ID) was invalid and that her son was still alive. He disappeared with a friend and they were buried together in Gacy's crawl space, and both were declared identified by the medical examiner's office. In addition, continued media attention perpetuated by two attorneys has focused on accusations of misidentification, possibility of additional victims, potential accomplices, and conspiracies of cover-up in spite of no documented evidence.

All ID materials were given to the attorneys so they could confirm the positive identification of this victim with any of their experts. In spite of the original ID documentation given to the attorneys, they proceeded to get a court order for the exhumation of the body of this questioned ID. Disregarding all the scientific evidence to date, the attorneys stated that the DNA comparison of the mother and victim's jaws "were not a match." The DNA results

were not made public and the laboratory would not comment to the Cook County Sheriff's Police Department. The attorneys stated publicly that the DNA comparison was made from the jaws, even though factually the jaws were not with the exhumed body.

Gacy, Mass Murder, ID Confirmation

F22 Examination of Dental Remains From a 19th-Century Burial Site

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The goal of this presentation is to describe dental restorations and prostheses recovered from a burial site dating from the 19th-century.

This presentation will impact the forensic science community by providing a historical perspective of dental practices and materials associated with this time period.

In the year 1851, the Erie County Almshouse was constructed on a 154-acre property. The building functioned at first as a poorhouse and later expanded to include a hospital and an insane asylum. In 1909, the property was sold to the University of Buffalo.

Those who perished while at the poorhouse were buried beginning at the eastern edge of the property, with later burials extending westward. When the University acquired the property, many of the graves were unmarked. They were also not relocated.

Although isolated remains had been found during minor construction over the years, it was not until a major infrastructure improvement project in 2012 that the possible extent of the cemetery started to become apparent. In the course of a salvage excavation, 480 burial locations were discovered, of which 383 contained human remains. It is estimated that there may be 3,000-5,000 remaining unexcavated gravesites within the property boundaries.

In a number of cases, dental restorative materials and prosthesis were found with the skeletal remains. Portable X-Ray Fluorescence (XRF) verified the composition of the metals present. The principal advantage of this technique is that it is non-destructive and rapid. This allowed verification of dental amalgam, gold foil restorations, platinum crowns, gold crowns, and precious metal bridgework.

An inspection of the metal prostheses suggested that the crowns were fashioned by a process called swaging. In this process, an impression of the patient's teeth would have been taken and a model made. The crowns would then be adapted to the teeth on the model by hammering the metal to fit. Both gold and platinum prostheses were present. These metals are malleable, making them desirable to use in this process, though platinum is more ductile than gold and would have been easier to work with. In the instance of fixed bridgework seen, the crowns were connected by solder.

Vulcanite dentures, complete and partial, were also discovered. The vulcanization process was discovered earlier in the 19th century; addition of sulfur and heat to natural rubber produced material capable of maintaining a precise but flexible shape. One of the first applications of this material was for denture bases. When cured, the material was dark orange in color. In order to make the material more esthetic, pink porcelain was adapted around the teeth to more naturally simulate gingival tissue. The teeth associated with these prostheses were made of white

porcelain. Aspects of the dentures recovered indicated that the practitioners at the time were capable of precise work.

While the availability of vulcanite dentures became widespread and affordable during the time period of the burials, the use of precious metals in dental applications must have been expensive for the patient. This burial site continues to reveal a fascinating picture of the life and times of those who passed through the doors of the Erie County Almshouse.

Though the Almshouse has been gone for more than 100 years, the main building of the original hospital is currently still in use. It has been modified and is now known as Hayes Hall and houses the State University of New York (SUNY) at Buffalo School of Architecture.

Forensic Science, Forensic Odontology, Dental Materials

F23 A Toddler's Remains Discovered: Science vs. Opinion vs. Hearsay — A Cold Case Reinvestigated

Bruce R. Wiley, DMD, 337 Greybull Avenue, PO Box 206, Greybull, WY 82426*

After attending this presentation, attendees will understand the necessity of constructively filtering hearsay in an investigation. In addition, the results of accepted scientific testing should supersede personal subjective opinion, but subjective opinion can necessitate further studies.

This presentation will impact the forensic science community by illuminating the fact that present forensic science knowledge is not able to answer every question that appears regarding human decomposition.

On the opening day of hunting season, October 15, 1988, two hunters discovered the skull of a child wedged in the crevice of a sandstone cliff in a remote area of the foothills of the Big Horn Mountains approximately 16 miles northeast of Greybull, Wyoming.

The authorities who were notified included the Big Horn County Sheriff and Coroner, who were part of an investigative team that went to the site to collect the remains. Both the Sheriff and Coroner reported smelling the odor of decomposing human flesh. The team recovered the fairly intact skull, although the zygomatic arches were missing bilaterally. Additionally, several long bones were found resting in a mass of what appeared to be adipose tissue. A leather strap, animal fur (antelope or deer), a piece of white loosely woven gauze cloth, and a flat piece of metal, which initially appeared to be a baby's spoon, were also recovered with the remains.

A noted anthropologist from the University of Wyoming was asked to examine the remains after their discovery. He was under the assumption that, from their condition, the remains were of recent origin, having only been at that location from a few months to a few years. Of particular concern was the apparent force which was used to push the body into the crevice, thus detaching the cheek bones.

Identification of the victim was impossible at that time. Therefore, the remains were separately placed in Zip-loc® bags and buried in a casket in an unmarked grave at a local cemetery. Interestingly, a local woman visited the grave after the burial.

In 2009, a court order allowed the remains to be exhumed for further analysis. Unfortunately, it was discovered that the seal of the casket, as well as the seals of the Zip-locs® had been breached by 20 years of groundwater, severely deteriorating the bones included in the remains.

The rumor from 1988 until present time was that a transient couple, living in a trailer home within a mile of the victim's discovery, "arrived with two children and left with only one." Many peculiar

behaviors of the family were reported during the investigation.

A coroner's inquest was opened regarding the case. Birth records were collected, registered sex offenders were interviewed, local citizens with knowledge of the incident were questioned, the lady who visited the unmarked grave was contacted, all historical records of oil production in that area at the time were obtained, DNA analysis was attempted on the unerupted teeth in the skull, and seemingly no stone was left unturned. The skull was sent to the National Center for Missing and Exploited Children to have a facial reconstruction completed. A primary incisor was delivered to the University of Texas Health Science Center San Antonio Dental School for analysis.

The investigative panel lost its support from the Big Horn County Sheriff's Department. The Wyoming Department of Criminal Investigation intervened and became the guidance for the panel, financially and from the aspect of law enforcement. Radiocarbon dating was conducted on the remaining long bones at the State's crime lab, leading to a rather surprising answer as to the identity of the victim.

This presentation will provide details of the investigation, which included an archaeological dig at the site of the victim's location, as well as the findings of the inquiry, as well as the results of which have led to still-unanswered questions in this query.

Cold Case, Hearsay, Investigation

F24 Forensics in Four Major Cities in the People's Republic of China

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WITHDRAWN

F25 Estimating Sex for African-Americans Using Diagonal Molar Measurements

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After attending this presentation, attendees will: (1) learn that methods developed on White populations are not accurate for estimating sex for African American populations; (2) learn how to apply discriminant function equations for the estimation of sex from human remains; and, (3) learn how to test discriminant functions, developed on a White Greek population and apply them to an African American population.

This presentation will impact the forensic science community by: (1) showing that this is unique research and has not been published previously for an African American population; (2) providing data that will help in the positive identification of unknown human skeletal remains in African American populations; (3) providing data which shows that, for forensic cases, methods employed for estimation of sex must be developed using population-specific samples; and, (4) following the *Daubert* and *Mohan* rulings that require forensic methods to be scientifically tested and reproducible, this study has done this for diagonal measurements of teeth in African American populations.

In forensic cases where skeletal remains are fragmented or incomplete, the teeth may be used in place of more traditional

bones for sex estimation. Using diagonal diameter molar measurements eliminates the need to measure between the interproximal spaces of the tooth, which can be difficult when the teeth are still located in the jaw.

The purpose of this study is to determine if the nine discriminant functions that were developed for a White Greek population will accurately estimate sex for an African American population. To do this, the current project assessed the degree of sexual dimorphism in permanent molars of African American skeletons using diagonal molar measurements of the crown and cervix, and then tested the effectiveness of each of the nine Greek discriminant functions for estimating sex using the African American skeletal sample.

In this research, 103 three individuals of African American ancestry were studied. Four diagonal diameter measurements were taken for each of the left mandibular and maxillary molars: mesiobuccal–distolingual crown diameter; mesiolingual–distobuccal crown diameter; mesiobuccal–distolingual cervical diameter; and, mesiolingual–distobuccal cervical diameter.

For all of the measurements, the percentage of sexual dimorphism yielded a positive percentage, indicating larger male dimensions for all molars. Statistical analyses revealed that the overall percentage of accuracy of the nine Greek discriminant functions for the entire African American sample (males and females combined) ranged between 47.9% and 55.9%. Function 5 (maxillary crown diameters) produced the highest percentage of accuracy (55.9%), while Function 2 (all maxillary molar diameters) produced the lowest percentage of accuracy (47.9%).

When estimating the sex of females only, the percentage of accuracy ranged between 0.0% and 16.3%. When estimating the sex for males only, the percentage of accuracy ranged between 95.5% and 100%. The mean measurement values for the African American females were larger than those for the Greek females, which may account for the increased likelihood of producing an estimation of male. The results suggest that the discriminant functions for the Greek population do not accurately estimate sex for an African American population.

Sex Determination, Diagonal Diameters, Molar Teeth

F26 Sex Determination Using Mesio-Distal Dimension of Permanent Maxillary Incisors and Canines in a Modern Chilean Population

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After attending this presentation, attendees will: (1) understand that dental methods developed on a Northern Indian population are not accurate for determining sex for Chilean populations; (2) learn how to apply discriminant function equations for determining sex from human remains; and, (3) learn how to test discriminant functions developed on a Northern Indian population and apply them to a Chilean population.

This presentation will impact the forensic science

community by: (1) providing unique research which has not been published previously for a Chilean population; (2) presenting data which shows that methods employed for determining sex from dentition must be developed on the population to which they will be applied, i.e., dental methods developed on a Northern Indian population are not accurate for determining sex for a Chilean population; and, (3) showing how a sexing method derived from dental measurements can be applied in subadult individuals ages 13 – 17 years. There are few studies that have focused on this age group, as the crown of the permanent incisors and canines develop at an early age. Methods derived from them will have a great impact on the study of juvenile skeletal remains.

After attending this presentation, attendees will understand the potential value of using the mesio-distal dimension of permanent maxillary incisors and canines to estimate the sex of unknown individuals. Because of the postmortem preservation and the fact that crown development is completed before puberty, teeth can be used to estimate the sex of pubescent individuals as well as in adults when other sex diagnostic elements of the skeleton are absent or badly preserved.

The purpose of this study was to determine if sexual dimorphism is present in the maxillary central and lateral incisors and maxillary canines of a modern Chilean population. A total of 205 molds (128 males and 177 females) were selected from the Departamento de Investigación del Instituto Nacional de Ortodoncia in Santiago, Chile, ranging from 13 to 33 years of age. Inclusion criteria consisted of individuals older than 13 years, presence of all six teeth, absence of caries, restorations, and crowding on the examined teeth. Patients exhibiting dento-maxillary anomalies such as cleft palate, shape and size teeth anomalies, and/or craniofacial syndromes were excluded from the sample. The maximum mesio-distal dimension was recorded for each of the six teeth investigated, measurements were taken from the vestibular aspect of the teeth, parallel to the occlusal plane, measuring the maximum dimension using a digital Vernier caliper accurate to 0.01mm.

The data was then subjected to statistical analysis; the mean, range, and standard deviation were calculated for each tooth, two-sample *t*-test was used to test for the statistical difference between means, and, therefore, to determine the presence or absence of sexual dimorphism. Pearson's correlation coefficient, *r*, was calculated to measure the effect size of the relationship between mesio-distal dimensions and sex. Canonical discriminant function coefficients were used to develop formulas to estimate sex from the teeth that exhibited sexual dimorphism.

The results of this investigation showed that although the mean measurements of the teeth were all greater in the male population, only statistically significant sexual dimorphism was present in both central incisors and canines, both left and right side. Pearson's correlation coefficient showed a medium effect size ($r = 3.7$) on the canines, whereas the central incisors only showed a small effect size ($r \leq 2.6$). Minimum values of 7.17mm and 7.33mm for the right and left canines and of 7.42mm and 7.45mm for the right and left central incisors were recorded on the male group. Maximum values of 9.2mm and 9.65mm for the right and left canines, and of 10.1mm and 10.55mm were recorded on the female group.

When examining the classification results to assess which tooth more accurately estimated sex, the right canine performed better with 78.5% correct female identification and 49.2% correct male identification. The left central incisor had the highest correct classification percentage for females (84.2%) but also had the lowest percentage for males (29.7%). The low correct classification for the males could be explained despite the significant values ($p \leq 0.001$) for sexual dimorphism – due to a major overlap of measurements between the sex groups, which is concordant with the effect size (Pearson's correlation coefficients) reported. Nevertheless, when

using the minimum dimensions of the canines found on men, 8% of the female sample could be correctly classified.

This research has shown that sexual dimorphism is present on the maxillary canines and central incisors but that the formulas derived from the mesio-distal measurements are of limited use because of the low correct classification of males. No previous research on teeth dimensions has been done on the Chilean population with the purpose of estimating sex. This study has contributed to the characterization of this population and could help in correctly identifying the sex in a small percentage of cases involving unknown individuals.

Dentition, Chilean Population, Sex Determination

F27 The Triple Test: Age Estimation Protocol for Unaccompanied Fugitives Developed at the Katholieke Universiteit Leuven, Belgium

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After attending this presentation, attendees will be informed on the age estimation protocol for unaccompanied fugitives used at the Katholieke Universiteit Leuven (KU Leuven), Belgium.

This presentation will impact the forensic science community by imparting knowledge that the Triple Test, the age estimation protocol for unaccompanied fugitives developed at the KU Leuven, Belgium, combines at least three medical tests based on clinical dental observations and dental or skeletal radiological registrations.

Most of the forensic dental age estimations need to be performed within the context of migration and asylum procedures. Based on the children's rights (Resolution 44/25, 1989), the protective status of a child has to be given to immigrating unaccompanied children. Related to immigrating people, the age of onset of maturity as defined in the country of arrival has to be considered. The authorities of the countries in which immigration is requested have the right to check the age of the applicant. Hence, medical age estimation tests are used. As an example, the age examination protocol for unaccompanied young fugitives developed at the KU Leuven and applied in Belgium is described. The protocol is mainly based on dental age estimation and integrates at least three gender-specific tests. Therefore, it was named the "Triple Test."

The Triple Test is performed after obtaining an informed consent from the applicant. It starts with a clinical dental examination to exclude diseases or syndromes possibly influencing tooth and skeletal development and also obtains a clinical impression of the dental age of the applicant. Consequently, the number of teeth, the amount of decay, stain, restorations, the positions of the periodontal attachment, the degree of attrition, especially on molars, and the dental occlusion are evaluated. The examiner registering the clinical impression is biased seeing and clinically examining the applicant. Therefore, all other parts of the Triple Test are also performed independently by a second examiner. In case the final results of both examiners are in disagreement, the tests are reconsidered until a full consensus is reached.

Next, a dental panoramic radiograph is taken and evaluated. If developing permanent teeth (except third molars) are observed, the age is estimated based on the registered developmental stages of the mandibular left permanent teeth using

the Willems *et al.* (2001) method. In case all permanent teeth (except third molars) are mature, the age is estimated based on the registered developmental stages of the available third molars, taking into consideration the missing third molars. Therefore, the Bayesian method developed by Thevissen *et al.* (2009) is used. The latter method allows one to calculate the probability of an applicant being older or younger than the age of maturity (18 years according to Belgium's law).

In addition to the panoramic radiograph, a hand/wrist radiograph of the non-handedness side is taken to verify the obtained dental test result. Therefore, the ossification of the hand/wrist bones, in particular, the ossification of the radius and ulna, is considered by using the Greulich and Pyle atlas (1959).

When the hand/wrist bones are mature, supplemental sterno-clavicular radiographs (frontal and oblique) are taken to observe the ossification of the medial part of both clavicles. Accordingly, the age is estimated based on the Schmeling *et al.* (2004) method. The evaluation of the clavicles allows the estimation of age, even when all available third molars are completely mature.

The Triple Test considers different age-related biologic variables and the obtained test results eventually define the estimated age more accurately, evaluate a wider age range, and obtain confirmation between test results. Due to biologic variance between people, scientifically unexplainable discrepancies between the test results can exist. In that situation, doubt about the estimated age exists, and Belgium law prescribes that the medical test delivering the youngest age result has to be taken into account (Wetgeving, 2002).

Age Estimation, Unaccompanied Fugitives, Triple Test

F28 Age Assessment Policies and Procedures for Undocumented Minors in Malaysia

Mohd Yusmialdil P. Mohd Yusof, MSc, Koning Albertlaan 216/001, Gent 9000, BELGIUM*

After attending this presentation, attendees will be informed on how age assessment procedures are taking place in Malaysia and the way they have been implemented according to Malaysian policies for undocumented minors.

This presentation will impact the forensic science community by providing a better understanding of Malaysian policies toward assessing the age of undocumented minors in regard to particular age groups, the incorporation of Islamic law (Sharia), the international bodies' involvement, and the role of local United Nations High Commissioner for Refugees (UNHCR). The issues between forensic and refugee-related matters pertaining to age assessment will be bridged and developed via interdisciplinary communication and collaboration.

Age assessment is particularly important to estimate the individual age and thus to render into which group the individual belongs. The particular age group varies and being 18 years of age remains the significant transitional year prior to becoming the age of majority. Consequently, according to the Islamic Family Law (Federal Territories) Act 1984 (Act 303) the minimum age for marriage is eighteen for men and sixteen for women. Life imprisonment is an alternative sentence for all crimes where the death penalty cannot be applied against children. In principle, children under 14 years of age cannot be sentenced to life imprisonment. However, this clause is voided if they are associated with people who possess firearms or explosives which are linked to terrorist acts. It is illegal for children under the age of 14 to work, but they are permitted to contribute to family business.

Malaysia is not a party to many of the key international

human rights instruments. Malaysia is neither a party to the 1951 Convention relating to the Status of Refugees nor to the 1967 Protocol relating to the Status of Refugees. As a result, by law Malaysia does not provide any specific formal protection to people who have fled their own country due to a fear of persecution on conventional grounds. However, Malaysia has, on humanitarian grounds, given temporary shelter to refugees until they can be repatriated to their homeland or sent to a third country for resettlement.

Due to this policy, all immigrants and asylum seekers are placed under the same flag and the responsibility for age assessment is given to the UNHCR local office. Malaysia, as well as the rest of the Southeast Asian countries except the Philippines, distinguishes only two main categories of migrants; namely, documented or "legal" migrants and undocumented or "illegal" migrants. The first category includes people who enter (and are allowed to stay) in Malaysia and who hold passports, visas, work permits and other valid documents, as required by the immigration legislation. The UNHCR has its own regulation on age assessment based on several international guidelines. The criminally charged undocumented minors face another hurdle in which they will be reprehended and referred to government-operated dental clinics for dental and skeletal age assessment.

In conclusion, age assessment performed by a health care provider or independent agency in Malaysia shows a variety of approach adopted from various policies and procedures. Standard operating procedure is a key answer to control the multiple avenues that may lead to age-assessment imprecision.

Age Assessment, Refugee, Undocumented Minors

F29 Dental Assessment of Alterations in Osseous Development and Its Impact on Post-Natal Age Estimation in Populations With Primary IGF-1 Deficiency

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After attending this presentation, attendees will be aware that considerations of growth deficiencies should be made when doing age estimation.

This presentation will impact the forensic science community by promoting the realization that there could be an error in age estimation if consideration is not made in regard to growth disturbances.

It has been observed among prepubertal children that were deficient in stature that a deficiency in Insulin-Like Growth Factor-1 (IGF-1D) is often a contributing etiology. Insulin-like Growth Factor (IGF) is synthesized in the liver and peripheral structures and circulates in a bound form to a number of proteins like IGF-binding protein-3 (binds 90% of IGF1 which circulates) for instance.^{1,2} The levels of IGF-1 fluctuate throughout life, peaking at the onset of puberty and decreasing in early adult life, and declining with the advancement of age, much like the levels of growth hormone do. In the pediatric population, the primary clinical display of this deficiency is short stature (subnormal linear growth), sub-growth velocity, or delayed onset of puberty which is associated with retardation of growth, when compared with age-similar population. Patients may be screened for developmental anomalies during their prepubescent years by assessing dental development both radiographically and clinically. It is conceivable that there is an underdiagnosed population of IGF-1D patients that has a clinical display of delayed growth and development yet no diagnosis from an endocrinology standpoint. Of this group of patients that are diagnosed, there is a subset of patients that has had treatment

to correct the IGF-1D condition which may also skew the dental vs. chronological age comparison and age estimation depending on when the diagnosis was made, when therapy was initiated and the duration of therapy. The other subset of patients are those that have received no therapy for the IGF-1D condition or have not been diagnosed at all. An accurate assessment with consideration made to delayed bone development when attempting to determine chronological age can be made with dental age determinants. Demirjian's method of age estimation for this population may not be the most reliable method of age estimation; however, other tools such as evaluating the cementum incremental lines, if possible, may be a more favorable approach.^{3,4} There are cultural and social economic considerations where diagnosis of this condition is not made because of the lack of diagnostic facility or a patient's non-access to care. From a forensic odontology standpoint, the examiner should take into consideration the display of un-erupted developed teeth, the presence of any non-exfoliated deciduous teeth and skeletal development and symmetry.

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IGF1-D, Age Estimation, Pediatric Growth Delay

F30 Age-at-Death Estimation of Historical Remains Using Dental Age Estimation and Skeletal Age Estimation Techniques: A Case Report

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After attending this presentation, attendees will understand some principles of age estimation using dental and skeletal methods.

This presentation will impact the forensic science community by illustrating the relative accuracy and use of various age-estimation techniques.

Introduction: Age-at-death estimation for subadults is often done using dental methods because dental development is very regular and is under fairly tight genetic regulation. The appearance and union of epiphyses also are useful in the morphological assessment of age-at-death estimation using existing reference standards. The purpose of this case report is to document the biological age-at-death of five subadult individuals from a 19th-century cemetery whose graves were exposed by flooding. Both dental age estimations and skeletal evaluation were used to assess the agreement of methods.

Methods: It was determined that four individuals fell into the category of adolescent (12-20 years of age) and one could be classified as a child (3-12 years of age) on the basis of physical evaluation and review of dental radiographs. All individuals had a diagnostic full-mouth series of radiographs for dental evaluation. The adolescents all showed evidence of eruption of permanent

teeth with developing third molars. The child was in a mixed dentition stage of development. The dental age was estimated for each case using atlas approaches.^{1,2} The adolescent cases were analyzed using the UT-Age Estimation database, based on third molar development stages.³ The child case was evaluated using age approaches as described by Moorrees *et al.* and Demirjian *et al.*^{4,5} The stage of union for epiphyseal fusion and primary ossification centers was recorded on a standard form, and age was estimated using standard data from reference data sets.

Examination of the adolescent cases all indicated that each individual except one was female based on *os coxae* criteria. The undetermined adolescent case and the child appeared to be too immature to make sex estimation. UT-Age tests for the adolescent cases were run using sex as female for the three cases, and the undetermined sex case was run as both male and female. Ancestry was set as African (Black) for all cases.

Results: All adolescent cases were found to be approximately 15 years of age using dental techniques, and the child had a median range of 7 years/9 months – 9 years/3 months for female and 8 years/2 months – 9 years/4 months for male using dental techniques. The skeletal techniques gave estimates of 15-20 years for three of the adolescents, and 10-15 years for the one with immature sexual differentiation. The child skeletal estimate was 5-10 years.

Discussion: There is generally good correlation between dental and skeletal age-at-death estimations within the scope of each method with significant overlap in estimation intervals. The UT-Age technique gave results that may be considered more precise and accurate when compared to other techniques because this technique is based on an individualized statistical interpretation of development with a median expression of age and 95% confidence intervals. The analysis of the child with the mixed dentition gave results that are all consistent with each other. The internal consistency between all techniques indicates that a good indication of biological age has been achieved.

Conclusions: (1) A variety of dental and skeletal age at death estimations for subadult individuals are consistent with each other within the limitations of each technique; and, (2) The UT-Age third molar assessment for adolescents gives the most precise and accurate age estimate when applicable.

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Age Estimation, Adolescent, Child

F31 Age Estimation Based on a 3D Cone Beam Computed Tomography (CBCT) Study of the Pulp Cavity and Hard Tissues of the Teeth for Forensic Purposes

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After attending this presentation, attendees will learn a quick method which can be of help in odontological age estimation procedures in the living and which is of striking value for its accuracy, simplicity, and usefulness.

This presentation will impact the forensic science community by providing the professional community with a quick and easy odontological method that is useful in current, often-requested age estimation procedures in the living.

Background: Age estimation is a mainstay of the reconstructive identification pathway of the living individual as well as of the dead, and, among the others, the odontological methods are considered to be quite reliable. In adults, the dental age can be estimated by the analysis of the progressive physiological and degenerative phenomena which affect the teeth and, among them, the secondary dentinal apposition. Along with the histo-pathological methods of dental age estimation, the recent introduction of the devices for the three-dimensional processing of the radiographic images of the teeth, such as the modern CBCT, allows the adoption of a non-invasive/destructive, reliable, and rather accurate method for dental age estimation.

Goal: The goal of the present research is to develop a non-invasive, conservative, reliable, accurate, and simple method of dental age estimation in the living by mean of the analysis of the volume decrease phenomena which affect the pulp cavity of the teeth. The examined method provides a specific software which allows the measurement of the volume of hard and soft dental tissues on the basis of the 3D radiographic images from the CBCT.

Materials and Methods: Two operators randomly selected and analyzed 295 CBCT radiographs of Caucasian subjects made for ordinary clinical purposes (Scanora® 3D dental cone beam unit). The sample of the CBCTs consists of 116 male individuals and 179 females in the age cohorts between 10 and 79 years. The image of the upper left central incisor has been extrapolated from the radiographs in DICOM file format using OnDemand 3D software. To measure the volume of the dental tissues involved, the images have been then elaborated with an innovative method of geometric approximation of the dental anatomy: the ratio "pulp cavity volume/dental hard tissues volume" has been obtained and then correlated with the age of the subject. An inter-rater agreement has been performed. The results have been statistically analyzed and a special but easily usable formula has been elaborated.

Results: The research needs to be implemented with the results from a larger number of exams before drawing final conclusions. The preliminary results are encouraging since the CBCT and the adopted method allow a precise and accurate measurement of the volume changes of dental tissues caused by aging. The research shows a high correlation between dentinal apposition and the calculated ratio, and the age of the individuals with respect to gender and age cohort. Moreover, the study of the volume instead of the linear or area measurement of the dental structures seems to be a promising approach, being the volume

less influenced by X-ray distortion. The easy calculation of the rate with the proposed method has taken no more than ten minutes each. Consequently, the method is quick and simple, and offers the best chance to reveal and to measure the correlation of such volume changes with the age of the subject, thus obtaining useful evidence to assess age of a living adult in forensic practice.

Conclusion: The presented method is a promising tool in the procedure for age estimation, permitted by the high technological level achieved by the currently available machines for the CBCT. Due to the correlation with age, the low-dose exposure to X-rays, the conservative and easy approach, time saved, and the economically irrelevant difference in costs between the CBCT and the Orthopantograph (OPG) exam, the adopted procedure can be considered technically reliable and affordable for forensic purposes.

Human Identification, Dental Age Estimation, Cone Beam Computed Tomography

F32 Using Combined Evidence From Dental Calcification and Wrist-Hand Bone Maturation to Assess Chronological Age

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After attending this presentation, attendees will be briefed on recent data on the comparison of dental and skeletal methods for age estimation and their accuracy. Three dental methods and three skeletal methods are adopted and their use explained.

This presentation will impact the forensic science community by describing the interdisciplinary results of the present research which bring an improvement in the procedures of age estimation in the living.

Background: The Age Estimation (AE) of children or adolescents is becoming one of the most relevant areas of forensic odontology, due to the increasing number of criminal cases involving young people, irregular immigration, and the many issues related to asylum seekers and adoptions. In the last several decades, researchers are ever more committed in searching reliable and accurate methods to assess chronological age of children and adolescents. Dental age estimated by tooth mineralization detected by Orthopantograph (OPG) and skeletal age assessed through the wrist-hand maturation evaluated by direct X-rays were found to be highly correlated with chronological age of the subject, thus resulting useful tools for accomplishing with AE demands. Dental methods and skeletal methods based on wrist-hand bones are largely experimented with and widely used for forensic purposes. According to research, very few studies compared dental and skeletal age on the same sample of children and examined a combination of dental and skeletal evidence for a possible improvements in the predictions of chronological age.

Goal: In the same sample of subjects, both dental age based on teeth mineralization and skeletal age through wrist-hand bones maturation was examined, comparing the reliability and the accuracy respectively of dental methods and skeletal methods. A possible combination of dental and skeletal evidence to improve the estimation of chronological age will be studied and pursued.

Material and Method: The sample consisted of 269 X-rays of left wrist-hand and 269 OPGs of Italian children aged between 6.93 years and 17.89 years of age (2530 and 6529 days) and was composed of 133 males and 136 females. The OPG and the wrist X-rays were taken on the same day. One forensic pathologist trained in AE and two trained forensic odontologists respectively and independently provided the skeletal age estimations and OPGs scoring. For bone AE, three methods were applied: Greulich and Pyle (GP) atlas-method and the Tanner-Whitehouse scoring-system, TW2, and TW3 version. Dental age was estimated according to three methods: Demirjian's original method (seven teeth), Demirjian's eight-teeth method, and Willems' method. No other information but the sex was available to the operators at the moment of age estimations. The intra-observer repeatability was tested asking the operators to repeat the estimations on 30 X-rays randomly selected from the entire sample after three weeks. A regression model was employed to combine skeletal and dental evidence for age prediction.

Results and Conclusions: The correlation index between chronological age and estimated age is high both for dental methods and for skeletal methods. On the basis of preliminary results, skeletal methods tend to over-estimate age more than dental methods. When combined, skeletal and dental evidence improve the prediction of age, thus statistical analysis is nearing completion.

Human Identification, Dental Age Estimation, Wrist-Hand Bone Age Estimation

F33 Age Estimation Standards for a Subadult Western Australian Population Using the Moorrees Method

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After attending this presentation, attendees will understand the principles and application of the age estimation method developed by Moorrees *et al.* utilizing the dentition of a subadult population.

This presentation will impact the forensic science community by describing the process of formulating age estimation standards specifically for a Western Australian population based on the Moorrees method. As a result of increasing global population mobility, contemporary age estimation standards are required and this study contributes to the body of information in this regard. This relatively uncomplicated method facilitates accurate chronological age estimation in subadults.

It is envisioned that, following this presentation, attendees will understand the principles and application of the age estimation method developed by Moorrees *et al.* utilizing the dentition of a subadult population.^{1,2} The Moorrees method is a non-invasive and simple approach and is the only standard to include data for both deciduous and permanent teeth from the same series of children.

Age estimation is a vital element of forensic dentistry and is an integral aspect of creating a biological profile. The dentition is widely considered the most reliable indicator of chronological age in subadults because tooth formation shows less variation due to nutritional and environmental factors compared to tooth emergence and skeletal growth. Radiological visualization of

Age Estimation, Dental Development, Subadult

F34 The Importance of Population-Specific Reference Standards in Dental Age Assessments

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dental developmental can thus aid in the accurate estimation of chronological age in forensic and archaeological cases. The visual atlas method of age estimation utilizes dental radiographs that show the distinct stages of tooth development, eruption, and exfoliation. The standards of Moorrees et al. are well-established methods of age estimation that use composite visual images of the developmental stages of individual teeth for each sex.^{1,2} For the estimation of age in subadult individuals, it is the method recommended by the American Society of Forensic Odontology (ASFO); however, it is based on the analysis of a North American population, and when applied to foreign populations, a loss of accuracy ensues. With increasing global migration, the most practical solution is to formulate contemporary population specific standards. Recently, a visual atlas was created using the Moorrees standards based on the analysis of dental radiographs from London.³ Such population-specific charts can lead to increased accuracy of chronological age estimations; however, the London atlas does not provide statistically quantified error rates for age estimations made using the diagrammatic representations.

The present study applies the Moorrees standards to a contemporary Western Australian population.^{1,2} The goal is to quantify dental maturation patterns in that population and create developmental standards for potential forensic application. A total of 380 digital orthopantomograms (185 female and 195 male) were analyzed; the stated age range was 4.0 to 25.0 years. In each orthopantomograph (OPG) the degree of development and eruption of the permanent teeth, and root resorption in the deciduous teeth, were assigned a numeric score to create a visual atlas (35 illustrations) encompassing individuals between 4-25 years of age. Dental development stages of the permanent teeth were regressed against chronological age; the Standard Error of Estimate (SEE) for the models based on individual teeth ranged from ± 1.124 years (right maxillary first premolar) to ± 2.197 years (right mandibular third molar) for the pooled sample. Multiple regression models improved prediction accuracy; the most accurate model (SEE of ± 1.092 years) incorporates all right maxillary and mandibular teeth (excluding third molars). The accuracy of all age estimation models was tested on a cross-validation sample of 50 OPGs.

The level of accuracy achieved in the present study is comparable to methods using skeletal markers, such as the ossification status of the medial clavicle epiphysis (SD ± 0.26 years to ± 4.21 years) and the radiographic examination of the hand (Greulich-Pyle method SD 0.6-1.1 years; Thiemann-Nitz method SD 0.2-1.2 years).^{4,5} This study represents the first-ever investigation of this method in a Western Australian population; the accuracy and relative simplicity of the method confirms its forensic applicability.

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After attending this presentation, attendees will be able to appreciate the importance of population reference standards in dental age assessments. This will be presented in the form of a research study that aimed to estimate the age of Chinese subjects using three different population-specific reference standards.

This presentation will impact the forensic science community by demonstrating the need for developing population standards for accurate age assessments. Attendees will further learn how to conduct age assessments through developing population-specific standards, which is a unique method developed by Dental Age Research London Information Group (DARLInG).

Age assessment has become an integral part of forensic medicine due to poor birth registration practices and the high number of asylum claims in major countries. Ethnic variation in physical maturity is an established phenomenon; hence accuracy of the estimated age frequently varies based on the reference data from which the original scores were obtained. To analyze the effect of ethnic specific standards, this study was designed to test the accuracy of age estimated from three different population standards. A total of 266 subjects constituting an equal number of males and females aged 2 to 21 years old, and of southern Chinese ethnicity were randomly chosen from patients of Prince Philip Dental Hospital, Hong Kong. Panoramic radiographs taken previously for clinical diagnostic purposes were scored according to Demirjian's classification of dental development stages. The Dental Age (DA) was estimated by obtaining mean ages of dental development from three datasets: French-Canadian, United Kingdom (UK) Caucasian, and Hong Kong (HK) Chinese. Following this, the difference between Dental Age and Chronological Age (CA) was calculated for each method (CA-DA). The overall mean age difference using the French-Canadian dataset was -0.62 years for males and -0.36 years for females, and 0.25 years for males and 0.23 years for females using the UK Caucasian reference data. The HK Chinese data estimated age as close as -0.02 years for both males and females. Statistical significance was set at $p < 0.01$ and the Pearson correlation analysis was conducted between the DA-CA values obtained from three datasets. Poor correlation was observed using the UK Caucasian ($r = 0.70$) and the French-Canadian ($r = -0.05$) datasets; however, in contrast, high correlation was observed in the HK Chinese ($r = -0.28$) dataset that was statistically significant ($p < 0.01$). It is concluded that the HK Chinese reference data accurately estimated the age of the HK samples; thus, emphasizing the need for population-specific data to ensure accurate age assessments.

Reference Data, Dental Age, Dental Maturity

F35 Comparison of the Diagnostic Accuracy of Cone Beam Computed Tomographs (CBCTs) and Orthopantomographs (OPGs): Clinical and Medicolegal Issues

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After attending this presentation, attendees will understand the evidence of the greater value of the CBCTs in the diagnostic resolution of pathological conditions in comparison to the OPGs.

This presentation will impact the forensic science community by suggesting the use of CBCT in some clinical circumstances as a consequence of the drastic reduction of the dose of exposure to X-rays permitted by the most recent radiographic devices and the reduction of the cost of a single examination.

Background: The OPG is the most widespread prescribed radiological examination. The introduction of digital imaging has allowed the reduction of the exposure dose and an important improvement in the quality of the images. Moreover, some three-dimensional radiological examinations, CBCTs among them, are used more often in dental diagnostic routines since they are a relatively inexpensive and easily accessible tool and provide for an accurate examination. Particularly, a CBCT scan allows one to accurately detect the site and dimension of oral pathological conditions which OPGs cannot definitely recognize, and implies very low radiation doses in comparison to the traditional CT. As such, the CBCT scan may become a recommended procedure in specific clinical conditions and omitting its use may raise medicolegal and ethical issues.

Goal: The goal of the present study is to evaluate the different diagnostic accuracies of CBCT compared with a digital OPG.

Materials and Methods: A total of 187 upper and lower dental arches CBCTs and OPGs, performed for ordinary clinical purposes in the same day, in the same radiology office, and with the same devices, have been submitted for the comparison to two general dentists. The operators examined the OPGs first and the CBCTs one week later, and listed in detail every detectable oral pathological condition or anomaly (transparencies, opacities, etc.). The difference in diagnostic accuracy between the two exams has been evaluated. The intra-operator variability has also been evaluated re-submitting 10% of the exams to the operators after two weeks. The inter-operator variability is also evaluated.

Results: The research is still in progress; however, it is already clear that the CBCT exam obtains a more accurate diagnostic evaluation of the oral clinical conditions than the OPG exam. The CBCT reveals more precisely the site and the actual dimensions of the pathological processes of the maxillary bones, which are only poorly or not at all detectable by the OPG. In most samples, at least one lesion not revealed by OPG is clearly shown by the CBCT, especially for periapical radiotransparencies of endodontically treated teeth. The most striking cases will be described and discussed. The ethical as well as the medicolegal implications will be discussed in terms of appropriateness of the two different radiological examinations, given their radiation dose, the costs and the difference in accuracy and efficacy in specific diagnostic procedures. The related medicolegal issues deserve further discussion to define the different radiological approaches and the possible implications in terms of standard of care.

Orthopantomograph, CBCT, Medicolegal Issues

F36 An Unusual Request

Peter F. Hampl, DDS, 7898 Greyhawk Avenue, Gig Harbor, WA 98335-4926*

After attending this presentation the attendees will gain information to be able to consider a new protocol in their forensic practice should they be requested to perform the unusual procedure of removing metal restorations.

This presentation will impact the forensic science community by developing a dialogue within the forensic dental community regarding this unusual request.

The value of precious metals has increased significantly in the last decade. There is increased awareness by the general public of the value of these precious metals. Many patients have significant quantities of these types of metals in the form of dental restorations. These can range from crowns, inlays, onlays, and even removable partial dentures or "grills." On a daily basis, there are requests on television, radio, and print media by various businesses to purchase precious metals. These requests are for any form of precious metals from jewelry to dental gold. The proprietors offer to evaluate the customer's "scrap" metal and offer immediate reimbursement.

Due to this widespread media blitz regarding precious metal values, family members aware of a decedent's dental history and restorations may be interested in recovering these items prior to interment or cremation. Removable items such as dentures and "grills" may be easy to recover. Cemented restorations require the skill of one familiar with the art of exodontia. A local funeral director requested the extraction of any teeth from a deceased that had "gold" restorations by the forensic dentist. This request was initiated by the wife of the deceased. The funeral director stated this request was clearly stated in the deceased's last will and testament. In addition the funeral director had obtained from the wife a signed addendum to the cremation authorization for the extraction of those teeth that contained precious metal restorations. In Washington, funeral directors/embalmers are not allowed to extract teeth from the deceased. Therefore, arrangements were made for the procedure to be performed at the crematorium. The details of the procedure performed will be presented. This presentation will allow the attendees to consider a protocol in their forensic practice should a request be made to perform this procedure. The purpose in presenting this case is to develop a dialogue within the forensic dental community regarding this unusual type of request: (1) is it ethical to perform this procedure?; (2) how many in the audience have performed this procedure?; (3) how many in the audience would refuse to perform this procedure?; and, (4) who better than the forensic dentist to perform this procedure?

Forensic Odontology, Ethics, Protocol

F37 Malpractice and Fraud in the First Degree

John P. Kenney, DDS, MS, 101 S Washington Street, Park Ridge, IL 60068-4290*

After attending this presentation, attendees will understand how proper documentation of patients, especially patients transferring into your practice, can be critical if malpractice or fraud by the previous practitioner is suspected or found.

This presentation will impact the forensic science community by providing a practical example of what can just "walk into your office" and how proper documentation, and compassionate patient care combined with forensic skills can work to correct one of the worst combined cases of substandard care and fraud.

In March of 2009, a new seven-year-old female patient (MM) with a toothache was referred to a private pediatric practice. What would follow was, to say the least, a journey to rectify the worst case of substandard care and fraud had experienced by this practitioner in 30+ years of pediatric and forensic dental practice.

The first of three siblings that were examined and treated MM, presented with multiple abscesses below restored teeth. A clinical exam and charting were completed, four periapical digital (Scan-X) X-rays were taken and read. Because of the acute infections present, the child was placed on amoxicillin for two days and was reappointed. The father was asked to obtain the treating dentist's records before his daughter's next visit, which he did. These records included three sets of bitewing X-rays all printed on the same 8½" x 11" photo sheet. Two sets of these X-rays were not properly dated, and were not correctly reflected in the dentist's record. According to the father, his children had been seen by the previous practice on a regular basis for over two years, and had numerous restorations completed by the practice owner or an associate. The records and X-rays were reviewed prior to commencing actual treatment on MM. Comparing those records to charting and films from the first visit were disconcerting. Due to the noted discrepancies, additional intraoral photos and a panoramic film were added to her record, prior to the five extractions. With the father's consent, the extracted teeth were preserved for any necessary future examination by insurance or other interested parties. A note to the tooth fairy also allowed an important common childhood experience to occur. Additional restorative and prosthetic treatment for MM was also necessary and completed over a period of three months. At the conclusion of treatment on MM and her two siblings (KM, CM), who also had significant issues with their care, insurance claims, and record continuity, individual letters were issued to the parents documenting findings and subsequent care. This was unusual, but this case was so far beyond routine occurrence, that the parents deserved documentation that they could easily understand, and take any action they felt appropriate. The parents, also patients of the practice in question, were referred to a general dentist for care and to correct any problems found with their treatment.

The documentation became critical to assist the State of Illinois Department of Financial and Professional Regulation to review and successfully prosecute this practitioner, ultimately resulting in a license revocation and the largest fine against a dental provider in the history of the State of Illinois.^{1,2} The practitioner had been cited three other times for bad record keeping, fraud and poor patient care. Both parents were insured, mom via excellent indemnity insurance and the dad via a high-benefit union dental plan. The practice owner, who of course is ultimately responsible for claims and what happens in their office, had billed both insurance plans as primary rather than following correct practice (the birthday rule familiar to any dental practitioner in the United States) to bill primary, await payment, and then bill secondary. In addition, the family had paid a significant amount of money as out-of-pocket costs. Poor dental care, fraud, and poor patient management all were present in these cases. Early in the investigation by the state, representatives of the U.S. Postal Inspector's Office and the Office of Inspector General of the U.S. Department of Labor initiated contact with the office because the father's union-provided dental insurance. Prosecution of this case by the U.S. government is pending.

Utilization of not only routine documentation skills for a dental practitioner, but forensic skills of dental aging, documents examination for continuity of content, detailed preparation of the IDFPF prosecuting attorneys, and expert witness testimony all were necessary to bring this case to a successful conclusion.

References:

1. Illinois Department of Financial and Professional Regulation, Division of Professional Regulation: Case 2010-xxxxx. Findings Of Fact, Conclusions Of Law And Recommendation To The Director, by the Illinois Board of Dentistry
2. Illinois Department of Financial and Professional Regulation, Division of Professional Regulation: Case 2010-xxxxx. Administrative Law Judge's Report And Recommendations

Dental Malpractice, Dental Fraud, Substandard Care

F38 Using Forensic Skills With Workers' Compensation and Personal Injury Cases

Cheri Lewis, DDS, 8500 Wilshire Boulevard, Ste 805, Beverly Hills, CA 90211-3106*

After attending this presentation, attendees will understand the principles involved in using forensic skills with workers' compensation and personal injury cases.

This presentation will impact the forensic science community by providing ways to use investigation skills in a systematic format with workers' compensation and personal injury cases. These skills can also be useful in professional malpractice cases and in working with state boards.

The qualifications required for working in these areas include coursework and passing examinations. One should have a comprehensive, up-to-date *curriculum vitae*. Methods are reviewed for obtaining cases and joining panels. One should charge appropriate fees for services provided and for interpreters when required.

The most common contested complaints are reviewed, including temporomandibular joint dysfunction, dental trauma, periodontal disease, and xerostomia-induced dental decay. Standardized examinations and testing for the complaints are discussed. It is important to have background material regarding other dentists involved with cases, including their *curricula vitae* whenever possible.

One should have a standardized approach for preparing for interviews of applicants/plaintiffs and for reviewing records. This includes developing a questionnaire for applicants/plaintiffs that should be mailed prior to appointments. The questionnaire should state that tape recording of interviews may occur. One should calculate the amount of time required to review records provided. One should assess inconsistencies in the records and determine how they impact the credibility of applicants/plaintiffs. Techniques exist for interviewing and reviewing records with the applicant/plaintiff. The elements of a comprehensive dental examination are reviewed including a temporomandibular joint examination and periodontal examination. The oral hygiene techniques of the applicant/plaintiff and their impact on dental health are discussed. Factors responsible for xerostomia including hormone changes, alcohol use, smoking, medications, and recreational drugs are reviewed. Also reviewed are the effects of obesity on dental health and the effects of smoking, medications, and drug use on temporomandibular joint dysfunction and dental decay.

Thorough analysis is given of the elements expected in a comprehensive report including billing, review of documents, history of injury, history of treatment, history of prior injury and/or disability, work history and job description, off work/recreational activities, family/social history, examination, diagnosis, permanent and stationary status, causation including reasons for opinions, subjective factors of disability, objective factors of disability, work restrictions, apportionment, and future medical/dental care

requirements. Intraoral photographs and medical/dental research can be useful.

Preparation for depositions and what to expect at them is discussed. For personal injury cases, information is required for educating attorneys, including preparation of questions to be asked opposing medical/dental professions. The preparation of documents and exhibits for trial is discussed as well as what to expect when providing expert testimony in a courtroom setting.

Investigation, Workers' Compensation, Personal Injury

F39 Case Studies of Failure to Diagnose Gingival Carcinoma: Was the Standard of Care Met?

John D. McDowell, DDS, University of CO, Dental Med, MS-F844, L26, Rm 130, 13065 E 17th Avenue, Aurora, CO 80045*

After attending this presentation, attendees will be able to recognize many of the common and less-common signs and symptoms associated with gingival carcinomas. Additionally, attendees will be able to understand the need for biopsies to differentiate between inflammatory, reactive, and neoplastic lesions of the gingival tissues. Attendees will also be able to understand the advantages of an early diagnosis over a late diagnosis and to recognize benefits associated with the development of a treatment plan that is favorable to patient outcome.

This presentation will impact the forensic science community by increasing the likelihood that health care providers will be able to diagnose gingival carcinoma in a timely manner. Although not common, gingival carcinoma (most often, squamous cell carcinoma) is not rare and has been reported as such in the literature for many decades. Although schools in the health sciences prepare future practitioners to evaluate and treat the most common pathologic conditions, these students (and eventually, practitioners) are also instructed in the less-common pathologic conditions which they might encounter. Health care providers must not ignore, minimize, or trivialize the uncommon conditions with which their patients might present. This is especially true when the diagnostic imperative (the entity the prudent clinician cannot overlook when practicing safely) is a malignancy. The literature indicates that gingival malignancies (especially squamous cell carcinoma) are not rare and cannot be ignored by health care practitioners as they develop a differential diagnosis for chronic gingival erythema/erythroleukoplakia or for those lesions that might appear to result from an inflammatory or reactive process. Several case studies utilizing patient histories, the analysis of physical examinations (including multiple clinical photographs), and test results leading to a diagnosis of gingival carcinoma will be presented. Clinician failures to use a proven diagnostic algorithm resulted in malpractice claims against those clinicians who failed to meet the standard of care.

Since most forensic odontologists are practicing dentists often diagnosing and treating several patients per day, these oral health care providers will encounter in their practices a wide range of pathologic conditions. Unfortunately, some of these pathologic conditions might be overlooked or misdiagnosed as non-pathologic conditions or variations of normal anatomy. Attending this presentation will increase the likelihood that health care providers will be able to diagnose gingival carcinoma consistent with the standards of care and in a timely manner.

When forensic dentists are providing case analysis or expert witness testimony, the diagnostic algorithm presented will assist them in assessing whether or not the standard of care has been met in the instant case.

Malpractice, Gingival Cancer, Diagnosis Through Biopsy

F40 Anatomy of a Bitemark Case With Comments on Unbiased Analysis, Terminology, Report Writing, and Second Opinions

Mark L. Bernstein, DDS, Univ of Louisville Sch of Dent, Dept of Diagnostic Sciences, Health Sciences Center, Louisville, KY 40292*

After attending this presentation, attendees will be able to evaluate two opposing opinions rendered in a patterned injury analysis and understand what the American Board of Forensic Odontology (ABFO) can do to continue to ensure objectivity in bitemark analysis.

This presentation will impact the forensic science community by showing that the current status of bitemark analysis remains prone to differing conclusions among experts. Improvement in ABFO guidelines and standards are needed to improve validity and reliability of the discipline.

Expert opinions drawn from bitemark analysis have been criticized in the National Academy of Sciences (NAS) Report as failing to apply the requisite scientific methodology that would ensure valid and reliable opinions. The ABFO has responded to this Report by imposing guidelines and standards with regard to terminology, techniques, and procedural safeguards. Yet cases still emerge where experts, presumably applying similar analysis, reach diametrically opposing views. These cases serve to legitimize the concerns of the NAS.

A defense attorney sought a forensic odontologist to review a case in which an opposing expert concluded that the defendant was the "probable biter" of a "lacerative" bitemark on the wrist of a homicide victim. Additionally, the victim was determined to be the "probable biter" of a bitemark in the defendant's arm. Both injuries were said to be prototypical bitemarks showing class and individual characteristics. A second opinion was consistent with this conclusion.

Upon review of the evidence and the prosecution's expert's report, the defense's consultant arrived at a different conclusion. This analysis opined that the injury on the decedent's wrist was "not a bitemark" by current terminology guidelines, failing to show anything more than two short linear red markings and no opposing arch. The injury on the accused was "suggestive of a bitemark" with consistent shape and size but without any defined individual tooth marks. Opposing the prosecution's expert opinion, the defense's consultant concluded that there was insufficient evidentiary value to comment relationship between the dentitions of either party to the injuries to which they were compared — let alone a confidence of "probable biter."

This study suggests that something other than a comparison between the anatomy of the dentition and the characteristics of the patterned injuries emboldened the opinion of the prosecution's expert. In this regard, commentary on bias, terminology, report writing and second opinions is examined.

In conclusion, this study recommends the re-examination of approved terminology, blinding of cases for analysis and accountability for second opinions.

Bitemark Analysis, Validity, Opposing Opinions

F41 *The Queen vs. J.A.A.*: Fresh Evidence Appeal in the Supreme Court of Canada Results in Granting of a New Trial in a Bitemark Case

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After attending this presentation, attendees will have a better understanding of: (1) why it is imperative that an expert in bitemark analysis examine all cases of purported bitemark injuries prior to cases entering the judicial system; (2) why the concept of a *Palmer* ruling and why post-trial introduction of bitemark expert evidence as “fresh evidence” is not, and should not be, necessarily automatically allowed; (3) how the forensic community must work cooperatively with prosecuting and defense counsel and police in instances where problematic evidence has been proffered; and, (4) how to make any submitted *curriculum vitae* as succinct and factually accurate as possible to prevent embarrassment to both the expert and the attorney that retained him/her.

This presentation will impact the forensic science community by providing law enforcement and children’s social services with the impetus to adequately document and investigate bitemark cases and to engage an appropriately trained expert to evaluate case material at the appropriate juncture — prior to the appellate process.

Original imaging evidence, which will be presented, will illustrate that the accused (J.A.A.) was charged with assault after allegedly sexually assaulting his ex-wife (S.A.), with whom he lived in the matrimonial home. It was alleged, in the course of the assault, that the complainant had bitten the accused across his finger as hard as she could. The accused was apprehended, photographed, and a police officer testified at trial that the mark on J.A.A.’s finger was a human bitemark. The accused was convicted by the judge who, in his ruling, used the bitemark evidence as corroboration of the complainant’s story. J.A.A. was, based partly on the bitemark evidence, sentenced to penitentiary time. On appeal, the prosecution obtained the opinion of a non-certified forensic odontologist that buttressed the policeman’s testimony stating that the marking was indeed a bite mark. Contact was made by the appellate law firm for an opinion on the case, and later, an examination of the credentials, and assistance for the defense in matters relating to the credentials of the prosecution expert. This was done prior to submission of the case to the Provincial Court of Appeals for admission of fresh evidence — the expert evaluation of the bitemark by the defense.

In the this jurisdiction, there is a three-step assessment of bitemark cases. The first step, after the examination of the material, is to determine whether the injury at issue is indeed a bitemark. It was the opinion of the prosecution expert that the injury was from a human tooth — most likely a cuspid tooth. The defense expert testified that the injury was not a bitemark. At the Provincial Court of Appeals, an argument by post-conviction defense counsel that the report of the expert stating that the marking was *not* a bitemark should be allowed as fresh evidence. The bar for hearing fresh evidence is high to prevent courts trying cases on multiple occasions. In the course of the cross-examination of the prosecution witness, it was evident that there were major issues with that person’s submitted *curriculum vitae*, resulting in a detailed cross-examination of over 90 minutes and ultimately withdrawal of

this expert’s report and testimony. Despite this, the appeal was denied in a 2:1 decision by the Provincial Court of Appeals and the case was ultimately heard by the Supreme Court of Canada. At the Supreme Court of Canada, the defense appealed that the new evidence — that what was originally thought to be a bitemark at trial was not — should be evaluated by the trier of fact and that the accused be permitted a new trial. In *Palmer vs. the Queen* (1980), the Supreme Court of Canada ruled that in order for fresh evidence to be allowed post-conviction, four criteria need to be considered: (1) the evidence should generally not be admitted if, by due diligence, it could have been adduced at trial; (2) the evidence must be relevant in the sense that it bears upon a decisive or potentially decisive issue in the trial; (3) the evidence must be credible in the sense that it is reasonably capable of belief; and, (4) it must be such that if believed it could reasonably, when taken with the other evidence adduced at trial, be expected to have affected the result. Before the court, the Crown prosecutor conceded that points 2 and 3 had been met so the appeal centered upon points 1 and 4.

The majority, led by Justice Charron, found that the defense had not met the due diligence criterion, but essentially agreed to excuse this, given that the Crown, too, had not introduced expert evidence regarding the bitemark. Justice Charron focused on the fourth criterion, setting up the trial judge’s decision as a “close call” between the competing testimonies of S.A. and J.A.A., with the presence of singular pieces of corroborative evidence, such as the bitemark, pushing the judge over the line beyond any reasonable doubt. Justice Charron maintained that the fresh evidence would remove vital corroborative evidence and undermine the credibility of S.A.’s testimony, particularly the segment about having bitten J.A.A. This was sufficient to reasonably expect that the defense expert evidence — that the marking on the skin of the accused — was *not* a bitemark — would affect J.A.A.’s verdict. On the basis of the Supreme Court ruling, a new trial was ordered. This trial never occurred and the defendant is a free man today, albeit substantially poorer and perhaps wiser. Attendees will see the documentary evidence on which the police “expert” and prosecution expert determined that the injury was a bitemark and on which the defense expert maintained that the injury was not a bitemark so that attendees may make their own determination.

Bitemark, Appellate, Supreme Court

F42 How Bitemark Analysis Can Assist the Courts

Richard H. Fixott, DDS, 3818 SW 21st, #202, Redmond, OR 97756*

After attending this presentation, attendees will understand the value of bitemark analysis as it impacts the legal system.

This presentation will impact the forensic science community by supporting the continued relevance of bitemark analysis in the courts despite calls for bitemark analysis to be deemed unscientific and invalid.

Bitemark testimony has been questioned by those in both the dental and legal professions. Some have stated that bitemark analysis is unscientific and should be excluded from legal proceedings and/or the courtroom. Two cases will be presented where bitemark analysis resulted in the accused admitting guilt when presented with the results of the analysis. The “confessions” saved considerable time and expense for the legal and law enforcement agencies.

Case 1: When a mother picked her daughter up from afternoon day-care, a bitemark on the child’s shoulder was noted. The child said one of the day-care workers bit her. The worker stated one of the other children made the bite. A photograph of the

injury and models of the worker's teeth were provided for analysis. The bite had few individual characteristics and no scale was present. The bitemark was consistent with an adult dentition, and all the children in the day care had only primary dentition. When confronted with the evidence, the worker admitted biting the child. She stated that the child was biting other children and she wanted to show her that it hurt. A plea was worked out and court time and costs were avoided.

Case 2: A 1½-year-old child, in joint custody, was left with the mother and her boyfriend. When the father picked up the child later in the day, injuries; including head trauma, a burn, and a bitemark were observed. The child was taken to the Emergency Room (ER) and examined. Photographs of the bite with a scale were taken. During the time the child was with the mom, the mother, boyfriend, and a 2-year-old child had access to the victim. The child was excluded due to arch size. Models and photos were taken of the two adults. A marked resemblance between the alignment of the boyfriend's teeth and the tooth marks on the child's shoulder was evident. Presented with the evidence, the man admitted biting the child and pled to the abuse. He received a ten- year sentence due to the severity of the other injuries.

These cases illustrate how bitemark analysis can assist the courts and law enforcement. Specifically, these cases allowed a plea to be entered that reduced court time and public expense.

Bitemarks, Jurisprudence, Odontology

F43 Democratization of Justice — A Dog Bite Case Gone Viral

Kenneth F. Cohm, DDS, 422 Teague Trail, Lady Lake, FL 32159*

After attending this presentation, attendees will gain an understanding of how social media can impact the outcome of a forensic case.

This presentation will impact the forensic science community by illustrating the increasing pervasiveness of media and public opinion involvement in forensic cases. Pressure brought to bear on public officials, law enforcement, the courts, and forensic experts can drive an investigation in many directions.

The internet is a pervasive and ubiquitous force in our lives. In particular, Facebook®, Twitter®, and YouTube® have the ability to shape public opinion and the direction of a case investigation. The power of e-opinion cannot, nor should be, ignored.

What started out as a casual play day for three young girls morphed into an "incident" that polarized a small town in Missouri and garnered national attention. It is yet another example of the transformative power of social media used as a platform to rally a cause. Close to 200,000 "experts" on Facebook® opined as to the fate of Phineas the dog. Although most expressed genuine concern for the dog, there was hearsay, inappropriate accusations, misinformation, and even threatened social disobedience to advance their cause.

So what was the issue? Several girls were playing in the yard when the family's Golden Labrador, Phineas, allegedly, accidentally bit one of the girls on the left side and dragged her a few feet. A playmate present confirmed this version of the story although the veracity of the witness testimony and the entire incident was later challenged.

The emergency room physician, based on the history provided, diagnosed the patterned injury as a dog bite consisting of a crush injury to the trunk, contusion, laceration, and puncture wound. Treatment consisted of an antibiotic ointment.

Shot records for the dog were unattainable. Consequently, Phineas was taken into custody by the code officer. City code defined a "vicious dog" as "any dog with a known propensity,

tendency, or disposition to attack unprovoked, to cause injury to, or otherwise threaten the safety of human beings or other animals." A hearing was required within 30 days and the mayor, with great controversy, appointed himself as the hearing examiner. Supposedly, there were two other incidents where Phineas bit both the victim in this case and her sister. The marks were described as "scrapes" and a "nibble." Phineas was declared a dangerous dog by the court and incarcerated.

Phineas went missing from custody at least once and was housed in multiple locations for months. A local attorney involved with The Lexus Project, a dog advocacy group in New York City, intervened on behalf of the family to prevent the dog from being euthanized. Supporters of Phineas from all over the world embraced social media on a "Save Phineas" Facebook® page. The fight for Phineas became contentious and was plagued with numerous accusations and defamatory comments and even personal threats. This controversy continued for well over a year.

Bitemark analysis was conducted independently by two experts, a forensic odontologist and an expert on dog bites and behavior. There was only one photograph available for analysis. The photo did not have a scale for resizing but other objects in the photo were used to scale the patterned injury 1:1. Both experts independently agreed that the patterned injury, even allowing for resizing error, was significantly smaller than Phineas' dentition. Based strictly on the limited evidence available, the large size discrepancy of the injury compared to the dentition, and with no credible corroboration of the event, allowed Phineas to be excluded as the biter.

In July, 2013, Phineas' attorney filed a motion in Circuit Court for relief from euthanizing Phineas for reasons of: (1) exclusion as the biter; (2) erroneous hearsay about previous bites; (3) misrepresentation of the severity of the wound due to altered photographs; (4) prejudicial hearing by the mayor; and; (5) the girls' family asked that Phineas be spared as he was not vicious.

This case is as much about social activism as forensics. There were many thousands of supporters trying to save Phineas from the gallows. Opinions for the most part shared genuine concern for the dog while uniting an army of arm chair experts in order to gain sympathy for the dog and his family.

The forensic community at large needs to be aware of the power of mass media public opinion influence in forensic cases. Social media activism requires social responsibility.

Dog Bite, Social Media, Facebook®

F44 A Discussion on Doubt in Bitemark Cases — Personal Experience

Robert E. Barsley, DDS, JD, LSU School of Dentistry, Oral Health Resources, Rm 5345, 1100 Florida Avenue, New Orleans, LA 70119*

After attending this presentation, attendees will better understand many of the elements that are present in bitemark testimony beyond the analysis and linkage to a suspect.

This presentation will impact the forensic science community by highlighting possible red flags that an expert should at least notice, if not heed.

This presentation will discuss four bitemark cases. A defense-hired expert was employed in two cases; as a prosecution expert in one case and, for lack of a better term, as a "community" expert in a fourth case. Each case will be discussed in terms of the sufficiency of the forensic dental evidence provided for analysis. The presentation will also discuss doubt in each of the cases from the perspective of the quality of the evidence, the completeness of the analysis (taking into account the methodologies available at the

time), the impact and understandability factor of the presentation in court, and the value of the opposing expert's case (should there have been one).

In the first case (which may be recognizable to many in the audience), the discussion will center primarily on the sufficiency of the evidence provided by law enforcement to the opposing experts. A secondary consideration in this case will be the quality and veracity of information covering the circumstances surrounding the events comprising the case. In the second case, which was appealed to the U.S. Supreme Court (*cert. denied*), but was later not retried after intervention by the Innocence Project, the discussion will center on the analysis itself and the lack of opposing forensic testimony at trial. The third case will discuss the serendipitous results reached when an administrative law judge decided to seek the "truth" by a conference between experts and opposing counsel in a potential child abuse case. The final case will again focus on the sufficiency of evidence and the use of forensic odontology to present a reasoned simulation of the events occurring during a "crime" well prior to the advent of video- and computer-aided evidence simulations.

Material from additional cases will be presented. It is hoped that the audience will gain a better appreciation of the myriad interactions surrounding a bitemark case — from the gathering and documentation of evidence, to the theories espoused by law enforcement and prosecuting attorneys, to how the evidence and analysis is presented at trial, as well as the influence (or lack thereof) occasioned by the presence (or lack) of an opposing forensic dental expert.

Bitemark, Reasonable Doubt, Expert Opinion

F45 Published Bitemark Research—Relevance in Real Bitemark Cases: Is It Helping to Better Understand Human Bitemarks in Living Skin?

Franklin D. Wright, DMD, 1055 Nimitzview Drive, Cincinnati, OH 45230*

After attending this presentation, attendees will better understand the current bitemark research models in relationship to real-world bitemark cases. This presentation will review published studies that discuss research findings related to bitemark analysis and comparison. The foundations and methodologies used will be discussed in relation to real cases involving bitemark-patterned injuries in living skin.

This presentation will impact the forensic science community by reviewing peer-reviewed published studies related to bitemark analysis and comparison in an attempt to open a discussion on the current direction of bitemark research and its relevance to real-world bitemark cases. This presentation will discuss peer reviewed articles that have been published in the recent past related to bitemark analysis and the relevance of the stated results of the studies to bitemark patterned injuries in living human skin. A review of the foundation and methods of the published studies will be presented and compared to examine how they relate to real world bitemark cases that occur in criminal events in society.

The studies focus on three main areas: recognition of bitemark-patterned injuries in living skin, transfer of the details of the dentition creating the bitemark to skin, and the uniqueness of the human dentition.

Recognition of patterned injuries in living skin is a critical first step if a patterned injury is to be investigated as a human bitemark. Guidelines published by the American Board of Forensic

Odontology offer an investigative pathway to identifying the characteristics of bitemarks in living skin as well as terminology to express the certainty that a patterned injury represents a bitemark. Recently published peer-reviewed research examined the ability of examiners to correctly identify patterned injuries as representing human bitemarks.

The scientific underpinnings of bitemark analysis and comparison are necessarily related to the ability of living human skin to record the individual tooth patterns in a biter's dentition such that the pattern is a representation of the biter's teeth. Recently published peer reviewed studies have examined laboratory inflicted "biting" patterns directed at examining pattern transfer to non-living human skin and pig skin.

The arrangement and uniqueness of the human dentition as it relates to human bitemarks in living skin has not been proven in large open-population studies. Recent peer-reviewed studies have examined the anterior human dentition using Procrustes analysis techniques to attempt to determine if the human dentition is unique.

Bitemark, Peer-Reviewed Research, Bitemark Research

F46 Expert Interpretation of Bitemark Injuries

Mark Page, PhD, University of Newcastle, Dept of Oral Health, Ourimbah, NSW, AUSTRALIA; Jane A. Taylor, PhD, University of Newcastle, 10 Chittaway Road, Ourimbah 2258, AUSTRALIA; and Matt Blenkin, MDSc, University of Newcastle, Ourimbah 2258, AUSTRALIA*

After attending this presentation, attendees will be able to describe some common sources of disagreement in bitemark injury interpretation.

This presentation will impact the forensic science community by pointing out that disagreement between experts is one of the most difficult situations faced by judges and juries when deciding whether to accept or reject expert evidence and, consequently, answer the ultimate question of guilt or innocence. Analysis of the sources of disagreement in bitemark injury interpretation is useful to the forensic odontology community in that it can direct appropriate interpretations along the bounds of what is scientifically valid as well as what views are consistently held by the expert community.

This study attempted to characterize the nature of disagreement amongst odontologists in determining the fundamental properties of suspected bitemark injuries. Fifteen odontologists were interviewed face-to-face and asked to freely comment on six images of supposed bitemarks. Interview participants were shown six images of bitemarks, all of them taken from actual casework presented to one odontology center in Australia. They were asked to assume that these were photographs given to them by an agency for their initial comments. No contextual information was given to participants, and they were free to express as much or as little opinion as they liked regarding the image before them.

Qualitative analysis using a grounded theory approach revealed that practitioner agreement was at best fair, with wide-ranging opinions on the origin, circumstance, and characteristics of the wound given for all six images. Expression of whether the injury represented a bitemark was generally expressed in one of five categories, ranging from weakly negative (probably not a bitemark) to strongly positive (definitely a bitemark), yet there was considerable variation in opinions about the same image in this regard. Other common areas of discrepancy included the supposed orientation of the bitemark, the significance of central ecchymosis associated with the injury, and whether or not the injury was suitable for further analysis. More experienced practitioners

(>10 years) agreed with each other less than those who had ten years or less experience in forensic odontology. There was no clear or consistent pattern to agreement or disagreement between any of the 15 odontologists; some practitioners whose comments were in agreement regarding one image were in complete disagreement concerning another, but nor was this consistently the case for any given odontologist pair.

Odontologists were also asked to rate the quality of the bitemark injury in accordance with a published significance and severity scale. Some issues noted with odontologists' application of this scale include its application in the absence of pictorial examples and its application to injuries that were older. The highest level of agreement in expression of the origin of the injury between odontologists was reached when the quality of the injury was poor, in that odontologists tended to agree most when there was uncertainty, as opposed to the lower level of agreement seen when more definitive opinions were expressed.

The differences in opinions can be at least partly accounted for by the inconsistent nature of approaches used by different practitioners in assessing bitemark evidence. Some practitioners used different criteria for assessing whether the injury was caused by teeth compared to other odontologists. The results of this study indicate that more definitive guidelines as to the assessment of bitemarks as patterned injuries should be developed in order to ensure the highest possible level of practitioner agreement.

Bitemark, Odontology, Forensic Science

F47 An Examination of Bitemark DNA Exoneration Cases: Past, Present, and Future Perspectives

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After attending this presentation, attendees will understand the role that bitemark evidence has played in DNA exoneration cases in the past 10–15 years. They will also understand some of the problems that have led to these exonérations and what can be done to minimize these problems in the future.

This presentation will impact the forensic science community by outlining how these past problems have affected the present status of the admissibility of bitemark evidence. It will also impact the future status of bitemark evidence in the court system, despite the previous legal precedents that established its admissibility.

Since 2000, there have been ten DNA exonérations in cases where bitemark evidence played a significant role in the prosecution of the case. An examination of these cases will be conducted with respect to a number of relevant factors including: (1) how many forensic odontologist(s) were involved in cases that resulted in exonérations; (2) the linkage opinion of the forensic odontologist(s) who compared the bitemark evidence to the defendant; and, (3) other evidence that may have contributed to the conviction of the defendant.

This information will be reviewed with respect to patterns that may emerge concerning the following: (1) were certain forensic odontologists involved in more than one of these cases?; (2) were the linkage opinions of these forensic odontologists the same or very similar?; (3) what role did the bitemark evidence play in the prosecution of the case?; and, (4) was there other evidence (aside from the bitemark evidence) that was presented at trial and what role did that evidence play in the conviction?

These issues will be discussed along with any patterns that emerge from the information collected. Any patterns associated with the aforementioned questions will be critiqued from

the perspective of what impact these patterns may have had on the outcome of the case. Although the introduction of other types of evidence may have played a role in the outcome of these cases, the perspectives about these cases will be limited to a critical review of the details of the bitemark evidence.

After a critical review of the bitemark evidence in these cases, there will be suggestions made as to proposals for minimizing these problems in the future and what the role of bitemark evidence may be, pending the outcome of present legal challenges to its admissibility.

Finally, the role of the American Board of Forensic Odontology (ABFO) will be examined with respect to what role it played in these cases, what is happening with the use of bitemark presently, and what actions the ABFO has taken to attempt to safeguard the value of bitemark evidence in the future.

Bitemark Evidence, Forensic Odontologist, DNA Exoneration

F48 U.S. Tooth Mark (Bitemark) Problems as Seen From Abroad

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After attending this presentation, attendees will acquire a more nuanced view on tooth mark examination, comparison, evaluation, and conclusion. Attendees may also acquire a better understanding of the scientific problems behind the assessment of a tooth mark case and a better understanding of the inquisitory system of experts in European courts.

This presentation will impact the forensic science community by providing a better understanding of tooth mark examination, comparison, evaluation, and conclusion. It may also improve the scientific research behind these analyses. Also, for the U.S. forensic odontologists, it will give a better understanding of the inquisitory system of experts.

For years, a debate has been going on in the American Academy of Forensic Sciences (AAFS) about bitemarks or tooth marks as some prefer to call it. The discussion is about the science of the examination of the tooth marks and if they should or should not be admissible as evidence in the court of law. This presentation will encourage discussion of some of the problems as seen from a European country (Norway) will be discussed.

One problem is that many American forensic odontologists seem to believe that it is their responsibility to tell if the person did in fact bite or not, which could impact whether they win or lose the case. It is therefore tempting to go too far.

Examination of tooth marks can be done in different ways. However, the important point is how many details we can see in the mark and if we find these details again in the teeth of the suspect. If we can, we must consider how likely it is that another person may see these details. That is the science and our area of expertise as we have an idea of how frequently the combination of details will occur. This is what the courts should be told.

It may not be so well known to U.S. forensic odontologists that the role of the expert witnesses is different in most European countries compared to Anglo-Saxon countries. The difference in roles between the adversary system in Anglo-Saxon countries and the inquisitory system in European countries will be discussed and also the possible influence that may have on the expert's testimony. In the inquisitory system, the role of the expert is to inform the court about the scientific value of the particular evidence and, in addition, about his own opinion regarding the case based on all facts. One can never be 100% sure that a particular person was the biter. An absolute conclusion may be binding for the court. In the European system it is left to the court to make the final decision based on a total evaluation of the evidence.

Admissibility is a specific Anglo-Saxon problem. In the inquisitory system, all evidence that the parties claim or the court asks for can be shown in the courtroom. It is up to the judge to decide how to weigh the evidence. A problem with not allowing tooth marks as evidence is that this could free a suspect. The so-called Innocence Project runs the risk of throwing out the baby with the washing water. Innocent suspects lose the possibility of proving they are right. The effect could be that innocent people may be sentenced.

Research in tooth marks has to some extent taken the wrong direction. Each tooth mark is different, and thus cannot be examined like a factory product or chemical test. Some marks have a lot of detail and may have a high evidentiary value. Other marks have few details and lower evidentiary value. The rate of failure is requested and can never be given as it will vary from case to case and from examiner to examiner. Thus to assess this, each mark and examiner should be exposed to a full scientific examination, which is, of course, impossible.

Quality assurance is important and a safeguard against wrong convictions. The International Organization of Forensic Odonto-Stomatology (IOFOS) has for many years advocated that two experts must agree on the conclusion and both sign the reports. Recently, the American Board of Forensic Odontology (ABFO) has also adopted this recommendation in tooth mark analysis.

Tooth Marks, Adversary System of Experts, Inquisitory System of Experts

F49 Superficial Forensic Evidence in a Child Abuse Case

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After attending this presentation, attendees will better understand the collaboration needed between odontology and other forensic disciplines.

This presentation will impact the forensic science community by increasing awareness of odontology's role and interaction with other disciplines.

Head trauma is a frequent occurrence in child abuse. The resultant neuronal body and axonal damage, vascular injuries with bleeding of a variety of types, and obstruction to the flow of spinal fluid may cause an impaired level of consciousness and focal neurological dysfunction as well as increased intracranial pressure. Secondary edema may further increase intracranial pressure. If severe, the elevated pressure may cause bulging of the fontanels and even separation of cranial bones along suture lines. The degree of initial damage and the development of secondary problems influence prognosis and ultimate outcome may not be known for months.

In addition to head trauma, there may be other injuries including intraocular hemorrhages, bone fractures, and a variety of superficial injuries as well as bitemarks. Bitemark evidence may be of use in excluding an assailant. A case of a comatose two-year-old African American female is presented where apparent bitemarks to the left chest were confirmed by the confession of the mother who committed the assault.

The mother initially claimed that the marks across the victim's left chest were bitemarks made by her three-year-old son, the third person in the room. However, the metric scale clearly showed a three centimeter arch width, which when compared to that of her son, might have refuted her claim.

The mother also claimed that the white mark on the victim's upper right chest was "ringworm" which the medical examiner

refuted. Eventually, she admitted that the mark was due to a belt buckle strike during the assault and that she had subsequently been putting skin lotion on it.

Since the mother confessed, bitemark evidence, including dental impressions, were unnecessary, and indeed such evidence is not always considered acceptable. However, all such findings should be noted and might have been useful if assistance in proof of the assailant's identity was required.

With the number of judicial overturns involving bitemarks such as those sponsored by The Innocence Project, some states have barred bitemark evidence from their courtrooms. Further, defense attorneys in that organization have scheduled seminars discussing methods for attacking such evidence. The 2009 National Academy of Sciences (NAS) Report raised a level of uncertainty in the legitimacy of bitemark evidence. Nevertheless, the case presented above may be used if not in helping identify the perpetrator at least to exclude the three-year-old in the room and in combination with other superficial evidence, elicit the mother's confession.

Odontology Section of American Academy of Forensic Sciences (AAFS) might consider its role in support and reliance on other AAFS disciplines.

Bitemarks, Child Abuse, Superficial Evidence

F50 Evaluation of Multiple Bitemarks Aids Law Enforcement: A Case Report

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After attending this presentation, attendees will understand the features of a bitemark used to aid law enforcement in a case of a small child with multiple pattern bruises. The presentation will describe how bitemarks can be used to help authorities direct the course of an investigation even though a specific biter was not identified.

This presentation will impact the forensic science community by demonstrating the value of bitemarks in cases where specific identification of the biter may not be the primary purpose for requesting evaluation of multiple pattern bruises suspected of being bitemarks.

This is a report of a case involving multiple bitemarks on a three-year-old girl. The girl had spent her day in day-care under the supervision of the day-care workers. Her mother picked her up as usual after work and brought the little girl home. The actions of the child were normal and the evening family dinner was uneventful. Following dinner, the mother began to prepare her daughter for a bath prior to bed. When she removed the girl's outer garments, the mother found multiple marks on the girl's back, shoulders, and torso. When the mother asked the little girl what had happened, she said that she had been bitten in day-care but was unable to tell her parents who bit her. Her parents immediately notified police. The police responded to the call and, upon seeing the condition of the child, reported a case of possible child abuse. Police photographs were taken of the lesions on the victim and the parents were questioned. The investigation led to the day-care where the little girl had spent the day. The day-care workers denied any knowledge of what the marks were or how they ended up on the little girl in their care.

In the process of the investigation, the case was referred to the forensic odontologist for evaluation. Copies of the original photographs were sent with specific questions that the authorities wanted addressed to help them establish a direction for the investigation. First, they needed confirmation that the pattern bruises on the girl were indeed bitemarks. Second, if the pattern

bruises were bitemarks, they wanted an opinion as to whether all the marks were made by one individual or if the marks were created by more than one individual. Lastly they wanted an opinion as to the age of the party or parties involved. In particular they wanted an opinion if the bitemarks were likely made by an adult or a child.

Evaluation began with a thorough observation of the photographs. They were of adequate quality with a standard 12-inch ruler present in many of the photographs. Additional evaluation of the marks included the use of Adobe® Photoshop® to remove a portion of the ruler and apply it to the image of the marks to determine intercuspid width. An overlay was made of one of the marks which was overlaid on the other marks, comparing arch size, shape, consistency of the width of the incisal edges of the anterior teeth, and the spaces between the individual teeth.

Based on the results of the evaluation, the following conclusions were made in response to the authority's questions. First, most of the marks on the child could be confirmed to be human bitemarks. Further, they all appeared to have been committed by one individual. Based on the size of the arch and the size of the teeth, the bites were committed by another child. These measurements were certainly too small for an adult and the appearance of primary teeth in the maxillary anterior segment eliminates even an adolescent as a possible perpetrator.

Based on the findings of the forensic odontologist, the authorities declined to further investigate this case or attempt to file criminal charges against the individual guilty of inflicting these bites.

Bitemark, Child, Multiple

F51 Two Cases With Closed Suspect Populations and High-Quality Bitemark Evidence

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After attending this presentation, attendees will understand how high-quality bitemark evidence can be used in the legal system to assist investigators, prosecutors, defenders, and triers of fact with cases that include patterned injuries suspected to be bitemarks. Two cases that involved violent abusive attacks to a young child and featuring physical evidence sufficient to permit both bitemark analysis and bitemark comparisons are presented. Considering that the two cases had similar backgrounds and evidence, similar results may seem likely; the actual outcomes may prove surprising.

This presentation will impact the forensic community by detailing two cases with bitemark evidence appropriate for conducting bitemark analysis and bitemark comparison. These two cases illustrate the use of the American Board of Forensic Odontology (ABFO) Bitemark Analysis and Comparison Decision Tree to assist the forensic odontologists in making the appropriate analysis and comparison decisions. The backgrounds, processes, procedures, and conclusions for these cases demonstrate how appropriate evidence from high-quality human bitemark evidence in the skin of living humans can be valuable in the search for the truth regarding the identification, inclusion, or exclusion (with reasonable medical, dental, or scientific certainty) of those individuals who become suspects in a bitemark case.

Although bitemark evidence has become the most controversial aspect of forensic odontology and one of the most challenged forensic science practices, the appropriate analysis and comparison of high-quality bitemark evidence remains valuable

for those seeking the truth. Historical reviews of past bitemark cases reported in the literature primarily involve cases corrupted by bias, incompetent analysis, or both. These cases, some of which resulted in the conviction of innocent persons, have been the most commonly *reported* cases and those reports have been used by some to support their beliefs that bitemark evidence should not be admissible in the legal system. Although some cases ended tragically for those actually innocent, very many more cases with bitemark evidence have been completed in which the scientific analysis of the bitemark evidence, as a part of the overall case evidence, has been a valuable part of the investigation and prosecution of those who inflict physical attacks and abuses on innocent victims.

This presentation provides actual case studies involving two victims of physical abuse with bitemarks. Following ABFO Bitemark Standards and Guidelines and using ABFO Bitemark Terminology and the ABFO Bitemark Analysis and Comparison Decision Tree, these two cases demonstrate the value of bitemark evidence.

Case 1: A three-year-old child was the subject of a violent abusive attack at home. Among other injuries were two patterned injuries judged to be human bitemarks. Sworn statements were taken from the two caregivers. Both confessed to being present when the attack occurred but denied being responsible for the attack. Photographic and trace swabbing evidence was collected from both bitemarks. Statements from the abused child were recorded. One of the two bitemarks was judged to be of high quality and evidentiary value and the other of good quality for analysis and comparison; seemingly, an ideal bitemark case scenario, but...

Case 2: A mother reported leaving her twenty-eight-month-old child at a friend's home. The friend later called the mother to report that her child was breaking out with a strange rash in multiple locations. Hospital personnel notified the police and child protective services that the child was a possible victim of abuse with multiple bitemarks. Later, an odontological evaluation confirmed multiple human bitemarks with varying evidentiary quality. One injury pattern was judged to be a human bitemark with high evidentiary value. Law enforcement and protective services investigators developed a population of possible abusers that included the mother, the friend, the friend's four-year-old child, and the friend's live-in boyfriend. The boyfriend became the prime suspect and was taken into custody. The subsequent bitemark analysis and bitemark comparisons became key elements of this case.

Proper forensic odontological evaluation of bitemarks with high evidentiary value can assist investigators in identifying, including, or excluding biters, with reasonable medical, dental, or scientific certainty in cases of abuse with bitemarks. Forensic Odontologists can provide testimony that will assist triers of fact to understand bitemark evidence. If all aspects of cases are managed appropriately by forensic experts, law enforcement investigators, prosecutors, defense attorneys, juries, and judges, guilty suspects should be appropriately dealt with and innocent suspects protected and absolved.

Bitemark Evidence, Bitemark Analysis, Bitemark Comparison

F52 Case Study: Morphometric Analysis of Multiple Bitemarks Observed on an Infant

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After attending this presentation, attendees will understand the interest of a thorough analytical process concerning

the morphometric analysis of bitemarks on the one hand, and the implementation of derived technical procedures, as far as the dental impression-taking is concerned, on a young child on the other hand.

This presentation will impact the forensic science community by bringing appropriate methodological solutions to children suspected of having committed bites. First, the methodology undertaken allowed a dental impression on a young child suspected of being the biter to be obtained. Secondly, the digitized pictures of the suspect's dental impressions were used for each of the unproven bitemarks by applying a specific protocol.

The methodology put in place for this experience presents a double interest: First, it explains the adjustment of a protocol of dental impression-taking on a young child suspected of having committed several bitemarks on an infant who was found dead. Second, this methodology describes a simple protocol of morphometric analysis of bitemarks by superimposition of the dental impressions of the alleged biting child on the different bitemarks found on the victim's body.

The examination of the infant proposed to count and locate the injured sites, to record the existence of bitemarks on the young victim, and to draw up the damage assessment. Photographs of marks observed on the victim were taken. No tissue sampling was made in order to respect the physical integrity of the infant.

For each of the marks observed on the infant's body, the following questions were raised: (1) is the injury a bitemark?; (2) is it a human bite?; (3) do the age and the appearance of the injury correspond with the alleged assault and the time when this assault had been committed?; (4) does the bite show usable characteristics?; and, (5) may these characteristics be compared with the teeth of the potential suspects. For each mark, an answer has been given by referring to the levels of trust of the guide of the American Board of Forensic Odontology (ABFO).

Because of the very young age of the suspect, the regular techniques regarding the taking of dental impressions on could not be applied. The choice was made to take impressions by a bite impression on a soft dental wax. Afterwards, these impressions were cast into silicone.

To be able to come to a decision, the verbal scale of EVERETT was used. It sets down the notion of hypotheses that must be studied in accordance with at least two alternatives: (1) the alleged attacker has committed the assault; and, (2) another person than the alleged attacker has committed the assault.

Conclusion: The bitemarks' characteristics have been compared with the suspect's teeth. A specific protocol necessary to taking dental impressions on a very young child had to be put in place. Another classic specific protocol was put in place for the comparison of the marks/impression data.

Eventually, all of these data were treated by referring to the verbal scale of EVERETT, which allowed putting forward a considered and logical conclusion.

Bitemark, Child Abuse, Postmortem Exam

F53 Using GNU Image Manipulation Program (GIMP) to Fabricate Bitemark Overlays

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After attending this presentation, attendees will understand the availability and utility of the GNU Image Manipulation Program (GIMP) and appreciate the simplicity of using it to fabricate bitemark overlays.

This presentation will impact the forensic science community by demonstrating a simple, low-cost method of

producing bitemark overlays.

Bitemark analysis is the process of studying a pattern to determine if it is a bite and then establishing the mechanism of its production. Food and skin are some of the common places bitemarks can be encountered. Human bitemarks on skin are the most common type of forensic bitemark case. Human bitemarks can be divided into adult and child groups.

The biter may be human, animal, or insect. Inanimate objects such as a pipe in cross-section or electrocardiogram pads may cause wounds that are very similar in size and appearance to human bitemarks.

Some foodstuffs are excellent in their ability to record an imprint of the dentition but over time lose dimensional fidelity due to dehydration. Human skin's ability to hold an imprint of the inflicting dentition is compromised by rebound and elasticity but is enhanced by the color shift due to contusion and to surface alteration. Bitemarks on living skin are lost to healing while those on a decedent are subject to decomposition.

Various methods exist to compare a dentition to a pattern. Currently, one of the most widely used methods is the overlay. Use of an overlay allows a graphical description of the relationship of the teeth to the wound. A computer with image manipulation software is used to fabricate the overlay. The method described here minimizes the amount of operator induced error, bias and subjectivity.

GNU Image Manipulation Program (GIMP) is an image retouching and editing tool and has been released as free and open source software. There are versions tailored for most operating systems including Microsoft® Windows®, OS X®, and Linux®.

GIMP has tools that are used for image retouching and editing, free-form drawing, resizing, cropping, photo-montages, converting between different image formats, and more specialized tasks. The developers and maintainers of GIMP strives for it to be a free, high-end software graphics application for the editing and creation of original images, photos, icons, graphical elements of web pages and art for user-interface elements. GIMP is available free and can be downloaded at <http://www.gimp.org/downloads/>. The download is 77MB and user's manual of 25MB is available in many languages. Ubiquitous help is available by using the Google® search tool. Once the download has been completed and GIMP has been installed on your computer, the GIMP icon can be double-clicked to start GIMP.

When GIMP is first started, three panels will be displayed: the main canvas; a toolbox panel and a panel with layers, undo; and, several other dialogs. A menu bar with access to several menus is displayed across the top of the main canvas.

Models of the dentition of interest are scanned and imported into GIMP. The models are scanned in the grayscale mode and the overlay is generated by using the threshold and stroke tools.

The overlay should be inscribed with pertinent information such as date, name of suspect, upper or lower, case number, etc. An inscription such as "this side down" is placed to ensure that the left-right orientation of the overlay is correct when in use. The image is now printed. A sheet of clear transparency paper is used for the actual overlay; the overlay image can also be printed on regular white paper for inclusion in a report. The image is saved as a GIMP XCF file to preserve the layers and the overlay is also exported as a JPG file.

Bitemark, Computers, Overlay

F54 Correction of a Type of Distortion Seen in Bitemark Photography: A Novel Algorithm With a Formal Proof

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The goals of this presentation are to inform odontologists and other practitioners who take photographs of patterned injuries on human skin of a new method to correct one type of distortion often seen in patterned injury photography and to inform practitioners of a method to improve the utility of patterned injury images with Type 1 distortion.

This presentation will impact the forensic science community by providing a reliable and repeatable method to rectify Type 1 distortion in bitemark and other patterned injury photographs that include the American Board of Forensic Odontology (ABFO) No. 2^o scale. The method is supported by a rigorous proof of correctness.

The technique allows the user to specify correspondence between four or more points in a digital photograph (or digitized film image) and a computer-generated model of the ABFO No. 2^o scale present in the photograph. This correspondence yields a mapping between the plane of the scale in the photograph and the plane of the film or image sensor in the camera.

The eight-parameter transformation between perspective planes is algebraically rearranged to yield two linear combinations of the eight parameters of the transformation. The mappings derived from user input are substituted into the linear combinations to create eight or more linear equations of the eight parameters of the transform. The parameters of the transformation are estimated using multiple linear regression. Once the transform is known, multiple practical applications are facilitated including rectification, measurement, and image comparison.

In contrast to prior methods, this technique can be completed in minutes and reliably rectifies this type of distortion. Images captured by as much as 80° from the optimal angle have been rectified. The eight-parameter transformation between perspective planes is an effective technique to rectify Type I distortion in patterned injury images utilized in forensic odontology.

Bitemarks, Patterned Injuries, Photographic Distortion

F55 A Coordinate Measuring Machine and 3D Bitemark Analysis

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The goal of this presentation is to introduce 3D into the bitemark analysis discipline. This might solve some issues and controversy in the discipline.

This presentation will impact the forensic science community by enabling odontologists to present a better visual and verbal explanation in the courtroom for evidential purposes.

Background: Child abuse is a major health threat and 38% of the victims are recorded as having bitemarks. Correlating a bitemark to a perpetrator is at present subjective, based on 2D superimposition. Bitemark analysis has to be an objective in order to be substantive evidence in court.

Goal: The principle goal of this study is to develop an objective, quantitative, and reproducible 3D technique using a coordinate measuring machine, Incise Dental Scanner (IDS), to correlate and compare suspect's dentition and bitemarks.

Method: Six post-graduate research students were recruited voluntarily to participate in this study. Each participant was asked to bite into a cheddar cheese block (20×40×20mm) with their front teeth. The impressions of the teeth and imprints were digitized by the IDS at a scanning speed of 500 points/minute and 0.1mm scanning interval. Cloud software was used to analyze the data, as it performs 3D free-form superposition and the differences of mismatch from subtracted images.

Results: In 3D, the match of the outline frame of the bitten edges with their corresponding models was demonstrated in the form a color contour map. The descriptive statistical difference between the two corresponding images revealed a high degree of fit with average differences ranging between -0.6 and 0.9µm and square root of mean between 11.1 and 11.7µm.

Conclusion: It was demonstrated that IDS and the "Cloud" software have the potential to give quantitative matching between bitemarks on hard substrates and suspect's teeth. It also gives a virtual 3-D image which eradicates any potential tampering or damage that would otherwise occur with traditional items of evidence. This enables odontologists to present a better visual and verbal explanation in court for evidential purpose.

Bitemark Analysis, 3D, Incise Dental Scanner

F56 The NAS Report, Forensic Odontology, and a Path Forward

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After attending this presentation, attendees will learn the basis of the National Academy of Sciences (NAS) Report, its findings as it relates to forensic odontology, and recommendations for the science going forward. Attendees will learn from some critical analysis of bitemark research as well as the limitations of the NAS Report as it applies to the criminal justice system.

This presentation will impact the forensic science community by exposing some of the specifics of the NAS Report as it relates to bitemark evidence that will impact the direction of research in this field. Competence in analyzing a pattern injury in both live and experimental bites will be enhanced so errors of the past will not be repeated.

The NAS Report was published in 2009 entitled, *Strengthening Forensic Science in the United States: A Path Forward* and authored in part by a "committee on identifying the needs of the forensic science community." This comprehensive white paper covered all the fields of forensic science and did a critical analysis with recommendations for improvement going forward. Specifically in the field of forensic odontology, the focus was on the area of bitemarks. At the NAS hearing in 2007, Dr. David Senn was the only presenter representing forensic odontology. He spoke specifically about bitemark evidence. The area of basic research in the forensic odontology field was covered with the acknowledgement that more needed to be done. To date, the majority of bitemark research has been conducted with the use of a mechanical device applying a "pseudo bitemark" on non-living human tissue (cadaver bites). While these findings as they apply to cadavers are accurate for a mechanical bite, the study is academic when applied to mechanical bites on living volunteers and very misleading when real teeth are used in a real life violent battery during an assault or homicide. The NAS Report divided the forensic science fields into two major categories.

The first area is “analytical” which has been referred to as hard science and includes fields such as DNA, chemical analysis of materials such as paint, gunshot residue, chemicals, materials such as fibers, fluids, serology, fire, and explosive analysis.

The second area is “pattern/experience” evidence and has been referred to as “soft evidence” and includes such things as fingerprints, fire arm examination, tool marks, bitemarks, blood stain patterns, handwriting, hair, impressions (tires, foot wear), etc.

The path forward for all the forensic fields as pointed out in the NAS Report includes, but is not limited to, the following: peer review of cases (supervisor review) before trial; proficiency testing on a regular basis; and, board certification by an accredited board. Indeed, the Report mandated board certification before one could be considered an expert and provide testimony at trial.

The American Board of Forensic Odontology (ABFO) has addressed these recommendations in whole and in part with the updating of their standards, guidelines, and recommendations for analysis, reporting, and recertification, specifically as it relates to bitemarks. The path forward for forensic odontology in the field of bitemark analysis will be researched on “real life” cases, such as cases that have been adjudicated and cases that have been proven by witness observation and peer review by appellate court decision in order to be on a sound scientific basis. Critical review by a second or even a third expert before an opinion is proffered is now part of the protocol. The area of recertification, proficiency testing, and improved requirements for board certification have and are being addressed. Improvements in the field of bitemark analysis and bitemark comparison have been addressed by the ABFO; the NAS recommendations have been and are being implemented, so the path forward is progressing.

Bitemarks, Odontology, Analysis

F57 Validation Studies in Odontology: Part Two

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After attending this presentation, attendees will have an understanding of progress in the validation of the science underlying a basic activity of odontology: Antemortem/Postmortem (AM/PM) radiographic matching.

This presentation will impact the forensic science community by demonstrating the commitment of odontologists to addressing the *Daubert* challenge, and by providing preliminary data on error rates in a basic activity of odontology.

Introduction: In an effort to address the challenges raised by the *Daubert* Trilogy and the subsequent National Academy of Sciences (NAS) Report of 2009, a three-pronged approach to validation studies in forensic odontology was adopted. The three activities of odontology being validated are: (1) skills in dental anatomy; (2) skills in AM/PM radiographic image matching; and, (3) skills in age estimation from radiographs.

This presentation reports on Part Two of the studies: skills in AM/PM radiographic image matching. Part one has been reported on elsewhere.

Materials and Methods: A major problem in designing validation studies is the integrity of the database utilized. The use of odontology casework files as *known* matches is problematical, as the cases have generally used only odontology techniques to confirm identity, without independent confirmation. This study utilized radiographs from real DVI operations where the odontology

results had been independently confirmed by DNA techniques in every case.

The study was placed online and invitations to participate sent to all known forensic odontology organizations internationally. Participants logged on, accepted the ethical guidelines, provided basic demographic data, and completed the study. University students who were not dentally trained also completed the study.

The format was that the participant was shown two images, one antemortem and one postmortem, and asked to decide if they represented the same individual. Fifty image pairs were shown. The participant was then asked to indicate their level of confidence in their decision, and to decide on a level of confidence for their decision using at least one of two scales: the American Board of Forensic Odontology (ABFO) scale and the International Criminal Police Organization (INTERPOL) scale. Feedback and correct answers were provided at the completion of the study.

Results: Preliminary results suggest that the validation study in its current format is difficult. Even qualified and experienced odontologists make some errors when asked to use a binary same/different response scale. However, qualified odontologists perform better than non-dentally trained participants, who perform at above chance levels but who are less accurate than odontologists. The presentation will expand on these results.

Discussion: This study has some limitations. The design of the validation study does not completely reflect odontology casework. Odontologists do not necessarily work alone; a decision on identity commonly relies on comparison of many radiographs rather than a single image; there is less time pressure in a real casework situation; there may be opportunity to retake postmortem radiographs in a real casework or DVI situation. Nevertheless, this study represents a preliminary approach to validation of one of the basic activities of odontology at an international level with a significant sample size, using a database which is robust.

Validation Study, Radiographic Image Matching, Odontology

F58 A Proposed Human Bitemark Classification

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After attending this presentation, attendees will have acquired information regarding a proposed human bitemark classification.

This presentation will impact the forensic science community by providing a simple, logical human bitemark classification based upon measurable attributes, type, and location of the bitemark injury.

There are few references to human bitemark classification. Proposals have revolved around the type of injury, its anatomical location, the manner in which it was caused or its evidentiary value. Pretty proposed a bitemark severity scale, while Souviron used variations in class and individual tooth characteristics and assessed them as to one of four classes.¹⁻²

The former have two things in common: (1) they are primarily qualitative measurements; and, (2) they are subjective in nature.

The 2009 National Academy of Sciences Report, *Strengthening Forensic Science in the United States: A Path Forward*, recommended the creation of the National Institute of Forensic Science (NIFS).³ This agency should promote, among others, “The development and establishment of quantifiable measures of the reliability and accuracy of forensic analyses.”

The present human bitemark classification reflects this need and is based upon identified measurable attributes

(quantitative value). Generally speaking, as the latter increases in number so does its evidentiary value (qualitative value). There are always exceptions, of course clothing and biter/recipient movement may interfere with the qualitative value of the bitemark, as examples. As the evidentiary value increases, so does its potential for comparison with a suspect dentition. Lastly, when both quantitative and qualitative values increase, there is a greater potential for identifying the biter. Two other measurable attributes are paramount for distinguishing primary from secondary human dentitions. These are arch widths and opening diameter.

There is a famous quote attributed to defense counsel Johnny Cochran: "If it doesn't fit, you must acquit." Thus, if you can't measure, you can't identify the teeth. If you can't identify the teeth, comparison is not possible. The current American Board of Forensic Odontology (ABFO) guidelines reflect this outlook in the diagram in the Diplomates Reference Manual Section III: Policies, Procedures, Guidelines & Standards February 23, 2013 edition.⁴ In essence, it states if one "can identify individual arches and individual teeth," you can proceed to bitemark analysis and, potentially, to bitemark comparison.

This presentation will impact the forensic science community by providing a simple logical human bitemark classification based upon measurable attributes, type, and location of the bitemark injury. The simple formula: 2/4/C/SOULDER refers to an identified human bitemark involving two maxillary teeth, four mandibular teeth, inflicted by a child's dentition on the recipient's shoulder. Another example, 0/0/A/BACK, indicates a diffuse bitemark where individual teeth cannot be identified or measured, made by an adult dentition (because of the arch widths and opening diameter of the bitemark), on the recipients back. The formula gives an immediate perception of the quantitative value of the bitemark, the biter's dentition (adult, mixed, child, unknown), and where it was inflicted.

References:

1. Pretty IA. Development and Validation of a Human Bitemark Severity and Significance Scale. *J Forensic Sci* 2007;52: (3):687-691.
2. Souviron RR. A Bitemark Classification That Makes Sense. *Proceedings of the American Academy of Forensic Sciences*; 2012, Atlanta, GA.
3. Committee on Identifying the Needs of the Forensic Sciences Community, National Research Council. 2009. *Strengthening forensic science in the United States: A path forward*. Washington, D.C.: National Academies Press.
4. American Board of Forensic Odontology, Inc. *Diplomates Reference Manual*, January 2013 Edition.

Human Bitemark Classification, Bitemark, Forensic Odontology

F59 High-Speed Videography of Simulated Human Bites on the Skin of Living Humans

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After attending this presentation, attendees will have a better understanding of the response of living human skin to bites with simulated human teeth.

This presentation impacts the forensic science community by facilitating the examination of the behavior of living human skin

during biting and the study of the features and characteristics seen in living human skin after the creation of a bitemark.

Human bitemarks have been studied in various media including living and cadaverous human and non-human tissues. There is disagreement among investigators whether human skin can record marks made by teeth with sufficient accuracy to reliably allow identification of biters. Bitemark evidence has consequently been subjected to increased scrutiny. To date, no study has demonstrated in detail the dynamic mechanism involved during the creation of a bitemark.

This study utilizes high-speed videography to document the action of appropriately articulated models of human teeth as they bite living human skin. The resulting videos allow study of the reactions of different subjects' skin during and immediately after controlled simulations of human bites. Serial still images demonstrate longer-term skin reactions and the nature and resolution of the patterns created by the human teeth models.

Although properly designed and performed studies using the skin of human cadavers are potentially illustrative, there are significant differences between the skin of living humans and cadavers. Disparities also exist between the skin of living humans and the skin of other living animals.

Consistent with Institutional Review Board (IRB) requirements for research performed on living human subjects, extreme care was given to the manner of recruitment and the treatment of subjects during the research. All consenting volunteers were over the age of 18. Care was given to ensure autonomy. Recruitment of volunteers was without pressure and/or coercion. Subjects were given full disclosure of all procedures and adverse effects of pain and bruising. All subjects signed a thorough informed consent including the ability to withdraw at any time without prejudice. No vulnerable populations were approached or used in any part of this project.

For this pilot study, ten volunteers were subjected to the simulated human bites using dental models of the upper and lower teeth of a single individual. The models were created using an epoxy resin (Primotec® Primopoly), a material that is close in hardness to human enamel. A force calibration device (Reid® Bite Reader) was used to measure the maximum-effort biting force of the single individual and to record the maxillary-mandibular biting relationship during biting. The models were mounted in that same maxillary-mandibular relationship with the anterior teeth in a slightly protrusive, end-to-end anterior relationship onto a mechanical biting apparatus that includes a modified Hanau adjustable articulator (Reynolds Controlled Bite Force Generator). The mechanical apparatus was calibrated using the Reid Bite Reader to produce the same biting force as the single individual's recorded earlier. The bite recipient subjects were blindfolded to inhibit anticipation. Each subject's right arm was placed into the biting apparatus and the biting sequence was initiated. Three digital video cameras were placed so as to record the bite event from different angles. The biting sequence was digitally recorded and, after biting, the subject's arm retained in the same position for ten minutes. After the initial ten minute period, single still images were taken every five minutes for a period of one hour. Additional still images of the areas bitten were recorded each day until the patterns were no longer visible.

The video data recorded at 60 frames per second are viewable in slow motion or normal speed allowing visualization and study of the living tissue response to trauma. The still images document the post-infliction responses of the skin of the ten subjects. These data may be used by forensic odontologists and others to augment the current understanding of the response of living human skin to injuries caused by teeth.

Bitemarks, Human Skin, High-Speed Videography

F60 A Follow-Up Study of Bitemark Characteristics in Live Human Subjects

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After attending this presentation, attendees will have a better understanding of the issues associated with performing bitemark research on live human subjects and the results of the latest attempt to gather research data.

This presentation will impact the forensic science community by reporting on the latest attempt to study the manifestation of bitemarks on live human subjects after altering the parameters of a preliminary study reported by Hermsen and Wilson.¹

Following Institutional Review Board (IRB) recertification, 22 young, healthy subjects volunteered for the project. The purpose of this study was to determine if changing the orientation of the dentition from centric relation to an incisive (end-to-end) relationship and changing the amount of tissue included in the bite would alter the results of the original study. In the previously reported study, the teeth had been mounted and the bite administered in centric relation. In addition, the bite had included the entire thickness of the forearm, from anterior to posterior (anatomical position). In this study, the incisors were aligned end-to-end and a smaller volume of tissue was engaged. As in the previous study, the bite was delivered by a device whose design was provided by Drs. Mary and Peter Bush and constructed by Dr. Eric Wilson. The device was fitted with a set of denture teeth provided by the prosthetics laboratory at Creighton University School of Dentistry and outfitted with a pressure sensor that could measure the amount of pressure being exerted by the bite. For this study, modification of the holes in the metal base to which the mandibular denture was attached allowed for the anterior/posterior reorientation of the teeth.

The intent was to deliver as much pressure as possible for five to six seconds, up to a maximum pressure determined in the previous study of 146 pounds for females and 235 pounds for males. The volunteers were all instructed to vocalize their desire to stop at any time during the procedure. There was also an observer who could abort the process at any time if it appeared that the volunteer was in distress or there appeared to be the possibility of skin tissue damage. Photographs were taken immediately following the bite and again at 24 hours.

In some respects, the results were similar to the previous study. As reported previously, there was significant variation in the manifestation of the mark at 24 hours and the females in the study tended to mark at lower pressures than the males. The time required for the skin indentations to disappear to the naked eye also stayed consistent, ranging from 20 minutes to over an hour. However, there were a number of significant differences between the studies. In the current study, none of the volunteers were able to withstand pressures approaching those in the previous study. The marks lasting 24 hours were obtained at significantly lower pressures with petechial hemorrhage and central ecchymosis a commonly observed feature. Among the volunteers was one of the volunteers from the previous study. Consistent with the results of the new group of volunteers, he marked at significantly lower pressure than in the study reported in 2012.

This study demonstrated one of the major issues in performing bitemark research on live humans: pain. About one-third of the volunteers exercised their option of stopping the bite prematurely due to discomfort. The other two-thirds were stopped by the observer as a precaution when it appeared the volunteer was in distress or there was the possibility of skin laceration or abrasion. When questioned, the participant who had volunteered

for the previous study admitted that he experienced significantly more discomfort even though the pressure exerted on his arm was significantly less than had been recorded previously. He described the previous study as producing pressure on his arm, like placing his arm in a vice, as opposed to the current study in which he felt the pressure but also, and more painfully, a pinching sensation. The end-to-end orientation of the teeth and the smaller tissue volume involved in the bite generated significant pinching of the tissue which is thought to account for the increased discomfort. The central ecchymosis often observed in bitemarks and more frequently observed in the current study than in the prior study, is also likely the result of the severe pinching of the tissue causing disruption of the underlying capillary beds. Despite the concerns of the observer at the time of the bite, none of the volunteers suffered abrasion of the tissue evident after 24 hours nor did any exhibit laceration of the tissue up to pressures as high as 135 pounds.

Reference:

1. Hermsen KP, Wilson E. A study of bitemark characteristics in live human subjects. Proceedings of the American Academy of Forensic Sciences; 2012. Atlanta, GA.

Bitemark, Human, Live

F61 A Comparison of Hollow Volume Overlays to Bitemarks in Vital Tissue When a Postural Change Is Effected

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After attending this presentation, attendees will become familiar with postural distortion of bitemarks and potential concerns of the forensic odontologist when presented with a bitemark for analysis.

This presentation will impact the forensic science community by adding to the scientific database of studies dealing with the coincidence of hollow volume overlays to bite indentations left in vital tissue.

It has been established in previous bitemark studies that distortion of skin itself, position of bite (relative to Langer lines), non-uniform height of the teeth, and other factors all contribute to the patterns which are left on skin and that these patterns are not necessarily identical to the overlays or the patterns left in inanimate recording media. It has also been shown that positional distortions of bitemarks occur.^{1,2}

Displacement studies (sequentially altering the horizontal and vertical relative position of teeth to their neighbors) have indicated that the features of displacement are more pronounced in the bitemark than in the actual dentitions, adding to the possible distortion. However, they have also indicated that displacement of 5mm between teeth enabled distinction between the dentitions.³

Odontologists are often presented with a bitemark discovered on a decedent during examination prior to autopsy. These marks, on a subject now supine, may not have been inflicted while the decedent was in the selfsame position, thus we are faced with distortion of the mark due to both the viscoelastic properties of skin and to postural changes. It is the goal of this study to compare marks left by two distinctly different sets of dental casts (hereafter referred to as Biter A and Biter B) in vital skin to the hollow volume overlays of said casts for coincidence when those marks are photographically recorded both in the actual three-dimensional attitude in which they were made (hereafter referred to as the spatial position), as well as in the anatomic position, the

position in which, very often, they are presented to the odontologist for evaluation.

Reproducible bitemark research, of necessity, has been carried out utilizing unembalmed human cadavers and casts of dentitions mounted on C-clamp-type locking pliers. This device is calibrated to deliver a known force upon closure. Actual bitemarks are generally caused during a struggle and rarely is the recipient of the wound passive. It is also reasonable to assume that said movement might create additional distortion. Another consideration is that the force of the musculature at the time of the bite is never known and, most assuredly, never universal. Thus, it is difficult, and may be impossible, to fully reconcile the reproducible science to the marks produced during violent encounters. This is an attempt to evaluate whether the distortions of bitemarks in human skin, caused by a variety of factors, can be so great as to allow for improper attribution of a bitemark when one is dealing with a very limited set of biters with dentitions which, even to the naked eye, are clearly dissimilar. That having been said, without a body of data to support our analyses, those analyses can, and perhaps should, be characterized as one's habit and intuition and not as logical conclusions based on research and the data generated by that research. This study wishes to contribute to that body of research.

Five "bites" were sequentially created on the subject's left arm using Biter A, the first four being parallel to the Langer lines, the fifth being perpendicular. Each bite was photographed immediately, with an American Board of Forensic Odontology (ABFO) scale present, in the spatial position and then in the anatomic position. The bites were also photographed in both positions at 10 minutes, 20 minutes, and 30 minutes. The initial photographs (time zero) were the only images utilized in this study. This process was repeated on the right arm of the subject, using Biter B. The respective hollow volume overlays, created as per ABFO guidelines, were compared to the bites in both the spatial and anatomic positions and vice-versa. The results will be discussed.

References:

1. Bush, M., Miller, R., Bush, P., Dorion, R. Biomechanical Factors In Human Dermal Bitemarks in a Cadaver Model J. Forensic Sci. 2009; 54 (1): 167-176
2. Miller, R., Bush, P., Dorion, R., Bush, M. Uniqueness of the Dentition as Impressed in Human Skin: A Cadaver Model J. Forensic Sci. 2009; 54 (4): 909-914
3. Holtkoetter, H., Bush, P., Sheets, H.D., Bush, M. Transfer of Dental Patterns to Human Skin AAFS 65th Annual Scientific Meeting 2013, Washington D.C. Presentation F29

Bitemark, Hollow Volume Overlay, Vital Tissue

F62 Tattoos: Identifiers and Cultural Reflections

Diane T.S. Penola, MA, 54 Fayson Lakes Road, Kinnelon, NJ 07405*

After attending this presentation, attendees will become familiar with the changing world of tattoos and the implications for victim identification.

This presentation will impact the forensic science community by drawing attention to the importance of gathering and documenting tattoo location and description of dental patients.

During the scientific session of the 2013 American Academy of Forensic Sciences (AAFS) meeting in Washington, D.C., Dr. James Schneider presented, *The History of Tattoos and Their Use as a Means of Identification*.¹ It was an effective presentation that brought to our attention the potential use of

tattoos as another means of positive identification. We are all intimately aware of the importance of dental X-rays and written records in victim identification. It makes perfect sense to have tattoo information a part of the written patient chart. Consider the identification team member who happens upon a dental record that specifies the presence of a tattoo with one big heart and two small ones, with three female names inscribed within. Finding a body that exhibits such a tattoo could conceivably be faster than comparing posterior root anatomy. Traditional dental antemortem and postmortem documentation and analysis, followed by comparison, would then be done to confirm the identification. The tattoo affords the convenience of a preliminary presumption, followed by a single in-depth comparison.

Dr. Schneider's presentation was impressive and caused a behavior change back in the office. Adult patients are now queried as to whether they have any tattoos. The results are in conflict with the prejudices of the baby-boomer generation. Tattoos were often considered a form of desecration found on motorcycle gang members, veterans of the Navy, and former prisoners. The demographics have changed dramatically. Middle-age females are one of the fastest growing groups of tattooed people.

Questioning patients about their status may be viewed as indelicate. When updating their medical history, a new question is asked about any existing tattoos. People often assume that the question references hepatitis or other "dirty needle" health issues. When the identification connection is made they may smile knowingly as they are somewhat familiar with forensic principles. Care should be taken to address the subject in general terms so as not to alarm them about their own possible demise. Explosive forces and body fragmentation are issues best left unsaid, even though they have impact on the usefulness of tattoos on the extremities versus the torso.

Blood centers have also changed their protocol with respect to donors with new tattoos. Depending on the state and the associated inspection requirements, individuals with new tattoos may not have to wait 12 months before being permitted to donate blood. This information can be used to initiate a conversation and encourage patients to become blood donors.

Tattoo shops have become big business and certain museums now view some tattoos as art. There is no doubt that we will be seeing increasing numbers of patients with tattoos. As they are often custom designed and quite unique in nature, their value as aids in positive identification must be recognized.

Reference:

1. Schneider, James C. "The History of Tattoos and Their Use as a Means of Identification" Proceedings of the 65th Annual AAFS scientific session, Washington, DC.

Tattoos, Victim Identification, Dental Records

F63 Superimposition 3D Skull/Picture of a Missing Person: A New Technique of Identification

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After attending this presentation, attendees will learn a

new technique of 3D surface acquisition and superimposition 3D skull using photogrammetry.

This presentation will impact the forensic science community by a standardized technique for personal identification based on 3D superimposition.

Much contemporary debate in forensic science concerns the alternative methods to approach at personal identification on skeletal remains. Emerging 3D surface acquisition technologies have recently introduced new skull scanning methodologies. Among passive methods, photogrammetry has proved to be particularly promising in the evaluation of skull morphology. Photogrammetry currently provides the most cost-effective 3D capturing system, being fast, inexpensive, non-invasive; the equipment necessary for acquisition is easily transportable and offers high reliability. The technique, indeed, was demonstrated to be suitable for capturing facial morphology for clinical and anthropological usage. This method of experimental research was aimed at developing an alternative to DNA analysis when this is not available. This study applied the technique of superimposition 3D skull/face pictures to 13 cases of identification of skeletal remains from unknown subjects: five women and eight men. In two cases, crossed superimpositions were applied; in the first between two skulls of female skeletal remains and in the second case between five skulls found in a cemetery mass grave. This technique involved four steps:

Preparatory Phase: A picture of missing person was analyzed and improved. Pictures with better view of missing person face landmarks were then chosen.

3D Acquisition Phase: A 3D photogrammetric avatar of the skull was created; this phase only required 4 photos simultaneously taken with a calibrated camera.

Superimposition Phase: This phase was the preparatory for the final step. The 3D skull was then carefully spatially oriented in the same position as in the photos and snapshots were taken. During the morphological analysis the snapshot of the skull was superimposed on the picture of the missing person's face picture by a specific software.

Metric Image Analysis: 3D skull than was carefully spatially oriented in the same position as in the photos, and snapshots were taken. To perform this step it was necessary to clearly recognize at least five landmarks on the skull using a suitable software. A 3D parameterized avatar of the skulls was created with a photogrammetric technique and photos of the missing person were selected and acquired. In the metric image analysis step, a quantitative comparison between the image of the missing person face and the snapshot obtained was carried out. Objective landmarks, as exocanthions, glabella, and subspinal point were marked on the 3D skull and on the missing person face to calculate the distance of the absolute points detected on the two images.

Results: The absolute and relative distances between the marked points, the perimeters, and the areas of the triangles, obtained by connecting the points, and the compactness indices were automatically calculated with a suitable program.

Conclusion: The morphologic phase revealed a full overlap between skulls and photos of disappeared persons. Metric phase revealed that correlation coefficients of this values, higher than 0.998-0.997, allow confirmation of the identification hypothesis.

Photogrammetry, Personal Identification, 3D Superimposition

F64 Dental Affections Related to Professional Activities in Senegal of a Forensic Odontology Interest

Khaliifa Dieng, DDS, BP6602 Dakar Etoile, Dakar 8622, SENEGAL*

After attending this presentation, attendees will have a better understanding of certain job- or trade-associated afflictions related to specific job tasks or activities.

This presentation will impact the forensic science community by providing a descriptive study of job-related lesions of the oral cavity and dentition and their relationship to potential pension benefits and victim identification.

Some bucco-dental affections relate to professional activities. In Senegal, for example, some craftsmen on the job use their mouth to hold small tools such as nails and screws, or even use their teeth to perform certain usual tasks. As such, the teeth gradually damage and induce specific dental infections which are likely to constitute true and reliable trademarks that can help retrace professional habits or perform estimative identification.

A sample of 670 individuals was selected from some trades in order to conduct a descriptive transversal study on the prevalence of dental lesions in relation to certain professional activities in Senegal. The study took about one year and the sample included 100 tailors, 100 tapestry makers, 100 cabinet makers, 100 shoe and talisman makers, 100 confectioners, 80 wrestlers, 40 masons, and 50 clarinetists.

The study took into account several parameters such as gender, age, and the number of years on the job. It specifically tried to determine the prevalence of those lesions relative to the trades according to the age and sex of the sampled population, as well as the number of years in the profession. Nevertheless, it should be mentioned, that natural, external, and internal causes should not be excluded.

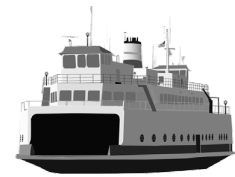
Having conducted interviews and thorough clinical examinations, the following prevalent dental lesions have been observed in relation with the selected trades: (1) there is a prevalence of 10.15% of cases of lesions of the free edges among the tailors, because they usually cut the threads with their teeth; (2) because of the heavy physical efforts, there is an abrasion prevalence of 5.22% among the wrestlers; (3) there is a prevalence of 2.09% among the masons, and 8.95% among the confectioners regarding caries of the snare of the teeth. For the masons it is due to the corrosiveness of cement but for the confectioners, it is due to the sugars; and, (4) there is a 10.25 % prevalence of edentulous, mobility, abrasion and periodontal cases noted among the clarinetists.

Admittedly, the sample does not include all the trades; however, establishing a relationship between some dental lesions and some professional activities is an interesting step for further forensic studies and industrial illnesses classification for the definition of pension benefits for certain industrial illnesses. The development and integration of such studies in forensic odontology activities will be helpful in terms of identification.

Dental Lesions, Professional Activities, Forensic Odontology



Pathology/Biology



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G1 Two Fatalities Due to the Use of Synthetic Cannabinoids Alone

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After attending this presentation, attendees will be aware of the rise in use of synthetic cannabinoids as an alternative drug. Attendees will understand that intoxication with synthetic cannabinoids is sufficient alone to cause death and will also understand the side effects and potential pathophysiology of deaths.

This presentation will impact the forensic science community by alerting the community of the potential for death due to synthetic cannabinoid intoxication in individuals without natural disease or other drugs contributing to their death.

The first case is an 18-year-old male with a past history of depression, bipolar disorder, and possible bath salt use. He had a recent history of telling his grandfather he was buying synthetic cannabinoids because he thought it was a safe, legal drug. He was last known to be alive the prior evening with complaints of not feeling well. He was found unresponsive in bed the following morning. Scene investigation revealed two empty silver packages suggestive of designer drugs, as well as white powder, and a leafy substance.

The second case is a 26-year-old male with a past history of hepatitis C infection and prior methamphetamine use. He reportedly stopped using methamphetamine 48 hours prior. He was last known to be alive when he went to bed. He was found unresponsive in bed two hours later.

The autopsy of case 1 revealed moderate pulmonary congestion and edema. The remaining vital organs, including the heart, did not exhibit significant anatomic or histopathologic change. The autopsy of case 2 revealed marked pulmonary edema. Intravascular birefringent foreign body material was histologically demonstrated in the lungs, consistent with a history of prior intravenous narcotism. Histologically, rare bronchi exhibited changes suggestive of a diagnosis of asthma. The liver had histologically demonstrable mild to moderate chronic inflammation, including eosinophils, but was free of fatty change and fibrosis. The remaining vital organs, including the heart, did not exhibit significant anatomic or histopathologic change.

In each case, femoral blood, heart blood, vitreous, urine, gastric contents, liver, and brain were submitted for toxicological analysis. Using gas chromatography with a flame ionization detector, two separate aliquots of femoral blood from each case tested negative for ethanol, acetone, isopropanol, and methanol. For each case, a 0.5mL aliquot of femoral blood was extracted with methanol and analyzed for nine drugs of abuse using enzyme

immunoassay. A 1mL aliquot of femoral blood was extracted using bicarbonate buffer (pH 11.0) and n-butyl acetate. The aliquot was then analyzed by Gas Chromatography/Mass Spectrometry (GC/MS) for more than 150 drugs. All screening and quantitation of femoral blood for synthetic cannabinoids were performed by an external laboratory using high-performance Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS). For case 1, analysis by LC/MS/MS revealed UR-144 at a concentration of 3.2ng/mL. No bath salts or other stimulant designer drugs were detected in his femoral blood, as analyzed by liquid chromatography/time-of-flight mass spectrometry. For case 2, analysis by LC/MS/MS revealed the presence of XLR-11 without quantitation. Vitreous electrolytes performed on case 2 were within normal postmortem limits.

The cause of death in each case was synthetic cannabinoid intoxication with the manner of death being accident. Documented side effects of synthetic cannabinoids include tachycardia, vomiting, central nervous system depression, seizure, hyperglycemia/hypoglycemia, hypokalemia, and arrhythmias. On scene, case 1 was found with vomit adjacent to his head. Myocardial infarction in living individuals has also been attributed to synthetic cannabinoid use. The above cases did not show any gross or histologic evidence of myocardial infarction, although this cannot be completely ruled out. Potential mechanisms of death in the above cases include central nervous system depression and arrhythmias.

Scene investigators and forensic pathologists should be aware that use of synthetic cannabinoids is increasing. These drugs are generally considered to be relatively safe and harmless by users. Currently, in many regions of the country, the drugs are also easily obtainable and relatively cheap. However, synthetic cannabinoids may not be detected on drug screens. Case 1 had a negative drug screen and the drug screen of case 2 detected cannabinoids. This case alerts pathologists that a positive cannabinoid on a drug screen may not simply mean the standard tetrahydrocannabinol. If synthetic cannabinoid use is suspected, the pathologist should notify the toxicology lab.

Forensic Pathology, Synthetic Cannabinoids, Intoxication

G2 Inconsistent Certification of Obesity in Natural Deaths With the Presence of Obesity-Related Comorbidities

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After attending this presentation, attendees will gain an understanding of the inconsistent inclusion of the term "obesity" on the death certificates of natural deaths determined to be due to obesity-related illnesses, such as coronary artery atherosclerosis, hypertensive heart disease, and diabetes. It will also be suggested that objective criteria, like the Body-Mass Index (BMI) of the deceased, are not routinely utilized during the process of death registration.

This presentation will impact the forensic science

community by heightening its awareness of the lack of standardization of the inclusion of obesity on the death certificate, a practice that has received relatively little debate during a period of rising obesity and obesity-related illness in the United States. With improved practices in the certification of obesity, the forensic science community can better inform the public of the number of deaths due to obesity.

Obesity is implicated as a significant risk factor for several types of chronic disease, and the rise in the number of obese persons in the United States has been accompanied by a rise in obesity-related morbidity and mortality.^{1,2} Although obesity is a known risk factor for many causes of natural death, obesity is listed as a cause of death relatively infrequently, as shown by a study in England.³ The forensic science community lacks evidence that a similar trend is occurring at medical examiner's and coroner's offices in the United States. Additional studies in this area are likely to lead to a standardized system of certifying obesity, which would improve the accuracy of the vital statistics that rely on information contained on the death certificate.

The death certificate records of the Hamilton County Coroner's Office (HCCO) in Cincinnati, Ohio, from the period of 2007 through 2008 that were ruled natural for manner of death were used for this study. Cases with coronary artery disease, hypertensive heart disease, myocardial infarction, pneumonia, sepsis, and diabetic coma or ketoacidosis indicated on part I of the death certificate were included in this study. Any cases with a mention of alcohol-related disease on the death certificate were excluded. Only cases with decedents between the ages of 25 and 69 years old were included in this study. Overall, 288 death certificates issued from the eight forensic pathologists on staff at the HCCO were inspected for the presence or absence of the term "obesity" in either part I or part II of the death certificate. The Body Mass Index (BMI) was calculated using the formula: (weight in kilograms)/(height in meters).²

Of the 288 death certificates used in this study, 125 were of decedents with a BMI of 30kg/m² or greater. The BMI range for the first to fifth quintiles were as follows: (1) 30.0-31.7kg/m²; (2) 31.7-33.9kg/m²; (3) 34.0-38.1kg/m²; (4) 38.2-42.5kg/m²; and, (5) 42.6-60.0kg/m². No death certificates in the first and second quintiles of BMI included "obesity." The term "obesity" was included on one death certificate in the third quintile, 11 in the fourth quintile, and 19 in the fifth quintile.

According to the study results, a BMI of 42.5kg/m², or the upper limits of the fourth quintile, serves as the threshold above which obesity is included on the death certificate in the majority of cases. All decedents in the fifth quintile have class III obesity, yet 20% of death certificates in this quintile did not have "obesity" listed on the death certificate. Moreover, less than 50% of the death certificates in the fourth quintile mention "obesity." The results suggest that death certificates of decedents with similar BMIs vary with regard to certification of obesity at the HCCO, and this is possibly due to under-utilization of BMI in death registration. A greater consistency in the utilization of the BMI would result in more death certificates containing the term "obesity" and, therefore, vital statistics that are based on those death certificates would more accurately depict the number of obesity-related deaths.

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Obesity, Death Certificate, Body Mass Index

G3 Postmortem Detection of 25I-NBOMe in Fluids and Tissues of a Young Man Who Fell Seven Stories to His Death

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After attending this presentation, attendees will be enlightened regarding the toxic and behavioral effects of an emerging class of "2C" designer drugs, which are N-benzyl phenylethylamine derivatives. Of particular interest in this case is the derivative 2-(4-iodo-2,5-dimethoxyphenyl)-N-((2-methoxyphenyl) methyl) ethanamine, also known as 25I-NBOMe.

This presentation will impact the forensic science community by providing practical information relevant to the postmortem evaluation of deaths associated with behavioral or hyperstimulation-type symptoms, particularly in cases where the initial postmortem toxicologic analysis is negative. This presentation will also present a novel drug of abuse that induces these types of symptoms.

In the last few years, these "2C" designer drugs have become easily obtainable over the internet, particularly 25I-NBOMe. These drugs are potent serotonin 5-HT_{2A} receptor agonists. The drugs are known for their perception-distorting and hallucinogenic properties. Street names for these drugs include 25I, INB-MeO, N-bomb, Smiles, Solaris, and Cimbi-5.

This study presents a case of a 19-year-old man who suffered multiple blunt traumatic injuries after he fell from the seventh floor of his apartment building. Prior to his death, he had ingested blotter paper containing "acid." His friends reported that after he ingested the drug, his behavior became bizarre, with paranoia being a predominant feature. Initial toxicological screens and analysis performed on body fluids and tissues by Gas Chromatography/Mass Spectrometry (GC/MS) and immunoassay were negative. Based on case history, a targeted analysis for Lysergic Acid Diethylamide (LSD) by immunoassay and Liquid Chromatography with Tandem Mass Spectrometry (LC/MS/MS) was also performed by a reference laboratory. No volatiles, drugs, or LSD were detected. Subsequently, the blotter paper from the stomach contents was analyzed by GC/MS; however, only bile salts were detected.

Based on negative findings and case history, law enforcement was contacted and the lead detective was able to obtain another piece of non-ingested blotter paper from the same source. Analysis of the non-ingested blotter paper by GC/MS identified the presence of 25I-NBOMe. A targeted analysis of 25I-NBOMe was then performed by a reference laboratory in Virginia.

At the reference laboratory, postmortem specimens were analyzed by High Performance Liquid Chromatography with Tandem Mass Spectrometry (HPLC/MS/MS). Toxicology findings for fluids based upon blood or urine calibrators were as follows: peripheral blood, 405pg/mL; heart blood, 410pg/mL; urine, 2.86ng/mL; and vitreous humor, 99pg/mL. Findings based upon the method of standard additions were: gastric contents, 7.1ug; bile, 14.3ng/mL; brain, 2.54ng/mL; and liver, 7.2ng/mL.

This study presents the circumstances of death, autopsy findings, and methods and results of toxicological analysis from a fatality associated with ingestion of the novel designer drug 25I-NBOMe. When confronted with negative postmortem toxicology results in fatalities associated with bizarre behavior

or symptoms attributable to hyperstimulation of the sympathetic nervous system, the forensic science community should have a high index of suspicion for intoxication with these types of drugs. Collaboration with law enforcement can be of great utility in these instances. Based on research of literature, this is the first presented postmortem case of 25I-NBOMe intoxication documented by toxicological analysis of tissues and body fluids.

25I-NBOMe, 5-HT_{2A} Receptor, Postmortem

G4 Fatal Metformin Overdose: A Case Report and Postmortem Biochemistry Contribution

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The goal of this presentation is to focus on a fatal case of lactic acidosis from metformin overdose in an Italian patient. Particularly, a complete forensic approach by means of clinical data collection, autopsy, microscopic examination, and toxicological and biochemical studies led to the conclusion that the cause of death was a terminal cardio-circulatory arrest resulting from lactic acidosis due to metformin intoxication.

This presentation will impact the forensic science community on the one hand by demonstrating how important a thorough forensic investigation is to reach the correct postmortem diagnosis and, consequently, to provide an adequate and high-quality service to the judicial authorities (in this case, medical liability profiles); on the other hand, it will impact society by showing how dangerous metformin administration could be for patients suffering from renal impairment and/or severely compromised oxygenation and tissue perfusion.

Metformin is an oral anti-hyperglycemic agent used in the management of type 2 diabetes mellitus. Metformin-Associated Lactic Acidosis (MALA) is clinically indistinguishable from other forms of lactic acidosis and generally characterized by unspecific symptoms. It is a well-described complication, with approximately 1 to 10 cases per 100,000 patient-years in patients chronically taking this type of drug. In the 2008 annual report of the American Association of Poison Control Centers (AAPCC), 6,538 cases of biguanide exposure were reported with 17 patients experiencing major adverse outcomes and eight deaths.¹ A five-year review of toxic exposures revealed that only 4,072 out of the nearly 11 million exposures involved metformin, with a total of nine deaths (0.2 % of all metformin-related exposures), 32 cases with life-threatening signs or symptoms and/or residual disability (0.8 %), and 187 cases with moderate clinical effects (4.6 %). These data suggest that lactic acidosis from metformin overdose occurs infrequently with therapeutic use and fatal cases, both accidental and intentional, are extremely rare in clinical practice. Even though lactic acidosis is a rare complication relative to the very widespread use of the drug, its possible occurrence still influences treatment strategies in type 2 diabetes mellitus, particularly in patients with impaired renal function.

This study describes a case involving a 70-year-old woman suffering from diabetes mellitus, obesity, and impaired renal function (creatinine 2.52mg/dl, corresponding to 223 μ mol/l, and urea nitrogen 121mg/dl, corresponding to 43.2mmol/l) who received metformin treatment (2,500mg/day). A medicolegal autopsy was ordered by the local inquiring authorities (public prosecutor) due to

the unclear circumstances of death as well as possible correlations between medical malpractice and the fatal outcome.

On the basis of internal examination (bilateral 100cc of pleural effusion, pulmonary edema, and multi-organ congestion), evaluation of histological specimens obtained from autopsy samples (Kimmelstiel-Wilson lesion of the kidneys), toxicological results (markedly increased blood metformin concentration accounting for 47.3 μ g/ml in femoral blood), and biochemical results (increased vitreous lactate — 30mmol/l — and increased blood and vitreous β -hydroxybutyrate — 110mg/dl=10,500 μ mol/l and 100mg/dl=9500 μ mol/l, respectively), it was possible to attribute the death to terminal cardio-circulatory arrest resulting from lactic acidosis due to metformin intoxication. Starvation ketoacidosis was also identified. Metformin accumulation was attributed to severely impaired renal function, which confirmed that drug prescription was absolutely contraindicated in this patient. Accordingly, the accused clinicians have been found guilty of manslaughter.

From a forensic point of view, such a case emphasizes the usefulness of performing exhaustive toxicology and biochemistry toward the more complete understanding of the pathophysiological mechanisms that may be involved in the death process. Moreover, from a clinical point of view, this presentation highlights that, although metformin is a highly effective, well-tolerated, and generally safe anti-hyperglycemic agent in the treatment of type 2 diabetes, a significant risk of toxicity has been associated with its use in certain types of patients.

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Metformin Overdose, Lactic Acidosis, Postmortem Biochemistry

G5 Retrospective Review of All-Terrain Vehicle-Related Fatalities in Puerto Rico

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The goal of this presentation is to describe All-Terrain Vehicle- (ATV) related fatalities evaluated during the period from January 2008 to July 2013 at the Puerto Rico Institute of Forensic Sciences.

This presentation will impact the forensic science community by describing 77 ATV-related deaths from a forensic perspective, focusing primarily on the types and mechanisms of injury.

ATVs are three- or four-wheel motorized vehicles intended for use by riders on off-road, non-paved surfaces. ATVs are a popular form of motorized recreation and are also used in a variety of occupational settings including agriculture, construction, and law enforcement. With their rising popularity, there was a corresponding increase in the incidence of injuries and deaths due to ATV accidents.

Seventy-seven cases of ATV-related deaths were received for postmortem examination during the period from January 2008 to July 2013 at the Puerto Rico Institute of Forensic Sciences. Of the 77 cases, 64 (83%) were males and 13 (17%) were females. In the majority of cases, the decedents were of White race (78%). Two fatalities (2.6%) corresponded to people younger than 16 years of age. The ages ranged from 16 to 20 years in 12 (15.6%) cases, from 21 to 30 years in 36 (46.8%) cases, from 31 to 40 years

in 21 (27.3%) cases, and in 6 (7.8%) cases, the decedents were older than 40 years of age. In 61 (79.2%) cases, the fatally injured person was the driver and in 15 (19.5%) cases, the passenger. In one case (1.3%), the driver of a motorcycle was hit by an ATV. In 63 (81.8%) cases, the accident occurred on a public paved road and 14 (18.2%) cases occurred in rural areas. In 76 (98.7%) of the 77 cases, the ATV was used for recreational purposes and in one case for work-related usage. The mechanism of injury included fall/ejection from the ATV in 61 (79.2%) cases, loss of stability and rollover in 3 (3.9%) cases, and collision with a stationary or moving object in 11 (14.3%) cases. In one case, the ATV and the driver were swept away by a river and in another case, an ATV hit a motorcyclist. In 24 (31.2%) cases, the death occurred at the scene, 19 (24.7%) of the injured died the same day under medical care, 24 (31.2%) died between the 1st and 5th day of hospitalization, and in 10 (13%) cases, the death occurred after the 5th day of the hospital stay. The injuries found at autopsy were divided into three groups. Head and neck injuries were present in 72 (93.5%) of the cases, thoraco-abdominal injuries were detected in 42 (54.5%) cases, and severe upper and/or lower extremity injuries occurred in 17 (22.1%) cases. Severe head trauma was the cause of death in 31 (40.3%) of the cases. In one case, the ATV driver drowned in a river. The blood alcohol level was less than 0.08%/weight in 14 (18.2%) cases and higher than 0.08%/weight in 11 (14.3%) of the cases. Among the illegal drugs, cannabinoids were detected in six cases, cocaine in one case, benzoyllecgonine in four cases, and opioids in one case. Alcohol and illegal drugs were detected together in six cases. In 50 (65%) cases, the toxicology came back negative for both alcohol and illicit drugs. Helmet and other security equipment usage were reported in only one case.

The results of this study show that most of the fatalities involved young male drivers. Passenger fatalities were mostly females. ATVs lack the general stability of other vehicles and are not meant to be driven on regular paved roads. This study shows that most of the accidents occurred on a public roadway during recreational use, which are recognized risk factors for ATV-related deaths. Fall/ejection from the ATV was the predominant mechanism of injury. This mechanism, in addition to the lack of use of a helmet, correlates with the most frequent injury found in this study. Head and neck injuries were present in more than 90% of the cases and severe head trauma was the cause of death in 40% of the cases. Head injuries are frequently fatal in ATV accidents, as demonstrated in these cases. Head injuries included subdural and subarachnoid hemorrhages, skull fractures, and brain contusions.

There is a lack of research regarding ATV-related fatalities from a forensic perspective. This study demonstrates a wide spectrum of injuries found at autopsy and correlates them with the mechanisms of injury.

ATV-Related Fatalities, Type of Injury, Mechanism of Injury

G6 A Different Truth at the Internal Examination: What You Can't Imagine at First Sight

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After attending this presentation, attendees will understand the extreme variability between the data obtained from the exterior

inspection of a dead body and those acquired as a result of an accurate autopsy examination.

This presentation will impact the forensic science community by serving as an example of the risk of a mistake occurring if, at the expert examination of an apparently straightforward case of death from "natural" causes, the related circumstances are taken for granted.

Presented here is the case of a young, homeless alcoholic whose body was found on a summer night, lying on his back at the entrance to a former garage, which was being used by squatters. At the site, after a first interview with the witnesses (friends) of the victim's last hours, the authorities were informed that the subject, addicted to alcohol for several years, had suddenly felt sick, thrown up, and fainted. The police officers at the scene presumed that these symptoms had been caused by a massive ingestion of alcohol. However, no traces of vomit were present, either on the victim's clothes or around the site. The witnesses did not provide any evidence revealing clinical elements that could suggest an ethylic coma or other hepatic complication generated by alcohol (rupture of esophageal varices and hematemesis). The exterior inspection of the body also yielded negative results, except for a faint, slightly abraded ecchymotic lesion at the level of the right costal margin. Because the cause of death was unclear, an autopsy investigation was scheduled.

As soon as the abdomen was opened, a massive hemoperitoneum was evident, due to many deep hepatic lacerations. It was then noted that the blended ecchymosis on the skin matched with fractures of three ribs on the right side, with associated hemorrhage. The decision was made to reinspect the victim's clothes and faint marks of a shoe on the right side of the T-shirt were recognized. Once these footprints had accurately been analyzed, it was ascertained, with the aid of the police officers, that they had been produced by a size 41 sneaker. The cause of death was determined to be massive laceration of the hepatic parenchyma due to trauma caused by kicking. Once the police had acquired these elements of proof, they were able to identify the killer by comparing footprints.

Too often, there is a risk that a presumably "simple" cause of death resulting from an apparently natural pathologic cause may lead to closing the file without first proceeding with an accurate autopsy inspection. Moreover, a negative exterior inspection of the cadaver may frequently hide lethal interior traumatic wounds. In all cases where, despite negative results of the first medical and forensic tests, the historic/circumstantial records seem uncertain, it is advisable to proceed with an autopsy to determine the actual cause of death.

Exterior Inspection, Accurate Autopsy, Faint Marks of a Shoe

G7 Blast Injury to Head and Neck: A Case of Suicide With a Homemade Explosive

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The goal of this presentation is to illustrate an unusual case of suicide with a homemade explosive.

This presentation will impact the forensic science community by demonstrating how this unusual method of suicide may result in potential exposure of first responders and medical examiner personnel to unique, biologically hazardous materials.

Introduction: Suicide is one of the most important public health issues and represents the 11th leading cause of death in the United States. Suicides comprise approximately 12% of the caseload of the Allegheny County Medical Examiner's Office in Pittsburgh, Pennsylvania. Suicide rates for this country have been relatively stable over the past decade, averaging approximately 10 per 100,000 populations. The most common method of suicide in the United States is the use of a firearm. Although decidedly uncommon, cases of non-terroristic suicide by explosives have been reported, raising issues of public safety outside those seen in more traditional cases of suicide.

Materials and Methods: Forensic investigators were called to the home of a 46-year-old Caucasian man found naked on the floor beside his bed in an inverted position with severe trauma localized to the occipital head and posterior neck. In addition, the left hand showed severe traumatic injury with partial amputation of fingers and extensive hand bone fractures. Investigation of the scene revealed innumerable minute fragments of paper/cardboard strewn across the room in the vicinity of the decedent's body. Investigators then found a box containing several apparent explosive devices of two different sizes, each of which appeared to be self-constructed. Each device consisted of a thick cardboard tube capped on both ends with a plastic sealant, and with a wick inserted into the body of the tube. Each tube was also filled with black finely granular material consistent with explosive flash powder. The body was taken for full postmortem examination and the flash powder from the devices was submitted for forensic laboratory analysis.

Laboratory and Autopsy Findings: Postmortem examination of the body showed an open wound of the occipital skull with associated brain trauma, which extended to involve the upper cervical spine and paraspinal soft tissues. This was consistent with an explosive-type blast injury. Laboratory analysis of the material inside the devices revealed flash powder (9.16 grams in the smaller devices, 22.95 grams in the larger). Further analysis of the material showed that it was comprised of a mixture of potassium perchlorate and aluminum powder. Taken together with the injuries of the left hand and recent threats of self-harm, it was determined that this was a case of suicide, committed by holding an explosive device to the back of the head and detonating the device.

Summary and Conclusions: Although rare, suicide by explosives does occur and such situations should be investigated with caution. Additional explosive materials may still remain on the premises and pose a potential explosive risk to first responders and investigative personnel. Furthermore, in cases such as this where significant amounts of residual flash powder may still be present, caution to avoid potential exposure and toxicity need to be taken. Perchlorate is known to interfere with normal thyroid function and, while not readily absorbed through the skin, can easily be absorbed through inhalation of powder particulate. Likewise, inhaled or otherwise ingested excess of aluminum can result in toxic effects to a variety of organ systems including the bones, bone marrow, and most notably, the central nervous system. Because of these concerns, the forensic community should make themselves aware of these rare explosive-related deaths as they have relevance for the health and well-being of the workers involved in investigations of these cases.

Explosive, Blast Injury, Suicide

G8 Case of Cyanide Intoxication With Subsequent Exhumation

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WITHDRAWN

G9 Sudden Unexpected Death in an Achondroplastic Dwarf

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After attending this presentation, attendees will be able to describe the anatomic features of achondroplastic dwarfism and list causes of sudden death in this patient population.

This presentation will impact the forensic science community by discussing a case identified as a pathologic substrate for sudden death in achondroplastic dwarfism, which may be more common than currently recognized. This case also emphasizes susceptibility to sudden death in this patient population and may assist in assigning the proper manner of death.

Achondroplastic dwarfism, the most common form of dwarfism, is a syndrome of characteristic anatomic findings resulting from a genetic mutation. The findings are predominantly skeletal and consist of an enlarged skull with a small skull base, a narrowed foramen magnum and spinal canal, short, flat, vertebral bodies, thickened intervertebral disks, shortened tubular bones and ribs with metaphyseal cupping, and shortened metacarpals and phalanges, among others.

This study presents the case of a 3-year-old boy with a past medical history significant for achondroplastic dwarfism. The decedent was at home with his two siblings in the care of his mother when he suffered a mild fall from a plastic step stool, causing him to fall backward and strike his head. Investigation revealed no history or suspicion of abuse.

Examination at autopsy revealed an enlarged cranium and short stature. A full skeletal survey confirmed the diagnosis of dwarfism and revealed no fractures. External examination revealed two green contusions on the forehead measuring 1.2 x 0.2cm and 2.0 x 0.4cm. There were no other external injuries. Autopsy examination was unremarkable aside from the achondroplasia, external trauma related to the fall, and a syrinx in the caudal medulla. No other traumatic lesions, recent or remote, were identified externally or internally.

Achondroplastic dwarfs are at increased risk of sudden death, although the anatomic basis for this is unclear and has been addressed only obliquely in the literature. One report by Mohindra et al suggests atlanto-occipital instability as a cause of death in these patients.¹ A study by Hecht et al found that there is an increased risk of sudden death in these patients which is attributable to brain stem compression in patients less than 4 years of age, while spinal stenosis contributed to the cause of death more frequently in older patients.² The findings in this case suggested

that skeletal abnormalities associated with achondroplasia, in particular a narrow foramen magnum, led to a syrinx in the lower medulla near vital cardiac and respiratory centers, increasing the likelihood that minimal trauma could lead to apnea and sudden death. Of note, lesions would go unnoticed unless sampled for histology. This case highlights the possibility that syringobulbia may be more common in achondroplasia than currently recognized and may represent a significant substrate for sudden death in these patients.

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Achondroplastic Dwarf, Sudden Death, Syringobulbia

G10 Sudden Cardiac Death in Young Adults: A Forensic Retrospective Study From 2007 to 2012

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After attending this presentation, attendees will understand that although smoking habits, alcohol consumption, and hypercholesterolemia were considered statistically significant when associated with coronary artery disease deaths in the group of "lifestyle risk factors," Body Mass Index (BMI) and diabetes were also present in the subjects of this study. Other causes of sudden cardiac death, such as primary arrhythmia or hypertrophic cardiomyopathy, should be identified in those who constitute risk patients, such as those who have a family history of sudden death. Thus, genetic screening plays an important role in the identification of genetic mutations implied in sudden cardiac death. Risk factors and the screening of genetic cardiovascular diseases shall in this way constitute a target of prevention.

This presentation will impact the forensic science community by illustrating that cardiovascular disease represents one of the most important public health problems in Western countries, with sudden cardiac death being the most common cause of death in adults.

Sudden cardiac death, the most common cause of death in adults under 65 years of age, accounts for an annual incidence between 180,000 and 250,000 cases per year in the United States (total population approximately 300 million) and between four and five million cases per year all over the world (total population approximately 6,540 billion).

Virmani *et al.* defines sudden cardiac death as a natural, non-violent, unexpected, and witnessed death within six hours of the onset of symptoms in an individual with a stable medical condition. This definition differs from that of the World Health Organization (WHO) which defines sudden cardiac death as a death occurring within 24 hours of the onset of symptoms. This period of time leads to the inclusion of well-established acute myocardial infarctions that should not be considered sudden. For unwitnessed deaths, both use the same definition, as a death occurring in an individual with a stable medical condition, who has been seen alive less than 24 hours before being found dead, and all non-cardiac causes were

excluded.

The objective of the present study is to determine the leading causes of sudden cardiac death in young adults in Northern Portugal between 2007 and 2012. This study includes 288 cases of adults aged 20-45 years of age who died suddenly of a cardiac cause and whose forensic autopsy was performed in the North Branch of the National Institute of Legal Medicine and Forensic Sciences between 2007 and 2012. Data included cause of death, forensic autopsy findings, and previous medical history as well as cardiovascular risk factors.

The mean age of the population was 37.36 years. Coronary artery disease was the leading cause of death found, representing 55.6% of all cases (92.5% of men and 7.5% of women). Females died mostly from a probable primary arrhythmia. Of those who died of coronary disease, 72 (25% of total cases) had histological evidence of a fatal acute myocardial infarction. Age, previous coronary disease, hypercholesterolemia, smoking habits, and alcohol consumption seem to be associated with an increased risk of sudden cardiac death by coronary artery disease. A total of 55.6% of deaths could be attributed to coronary artery disease.

Coronary artery disease presents as a silent devastating disease whose first manifestation can be sudden death. This study reinforces coronary artery disease as the main cause of sudden death in young adults and the leading cause of death in men, in whom the risk of coronary artery-related death is higher than in women. Older age was also a predictor of coronary artery disease deaths. Differently, in women, a probable arrhythmogenic cause of death was suggested in 51.2% of cases.

Cross-sectional severe luminal narrowing of coronary arteries was found in a large number of cases, with acute myocardial infarction a tragic consequence of that stenosis. However, silent infarction and chronic ischemic cardiomyopathy verified in some cases were also a consequence of different degrees of narrowing. The findings of this research suggest the need to reduce cardiovascular risk factors, as well as prevention and earlier detection of atherosclerosis, even in younger individuals. Although smoking habits, alcohol consumption, and hypercholesterolemia were considered statistically significant associated to coronary artery disease deaths in the group of "lifestyle risk factors", BMI and diabetes were also present in the subjects of this study. Other causes of sudden cardiac death, such as primary arrhythmia or hypertrophic cardiomyopathy, should be identified in those who constitute at-risk patients, such as those who have a family history of sudden death. Thus, genetic screening plays an important role in the identification of genetic mutations implied in sudden cardiac death. Risk factor reduction and screening for genetic cardiovascular diseases are methods for preventing sudden cardiac deaths.

Sudden Cardiac Death, Forensic Autopsy, Adults

G11 The Application of the Multi-Phase Postmortem CT-Angiography (MPMCTA) in a Rare Case of Spontaneous Rupture of the Right Common Carotid Artery in a Young Man

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The goal of this presentation is to analyze the value of the Multi-Phase Postmortem Computed Tomography- (CT) Angiography (MPMCTA) in cases of sudden and unexpected death related to cardiovascular pathologies.

This presentation will impact the forensic science community by demonstrating the necessity of a postmortem radiological examination and performing CT-angiography, especially in cases of sudden and unexpected death related to cardiovascular pathologies.

MPMCTA is a standardized technique consisting a native non-contrast CT scan, followed by at least three angiographic phases (arterial, venous, and dynamic phase). The angiographic phases perfuse the vascular system with an oily contrast agent mixed with paraffin oil, infused through intravascular accesses in the femoral artery and vein. The highly radiopaque iodized oil is transported in paraffin oil through the vascular system analogous to the delivery of contrast agent by the circulating blood in clinical angiography. The three phases (arterial, venous, and dynamic) allow complete opacification of the vascular system such that small vascular lesions or abnormalities can be identified as the exact source of bleeding, which could potentially be missed during the autopsy. Indeed, angiography could reveal more vascular pathologies than conventional autopsy and make it possible to identify the type of hemorrhages (arterial or venous) and their sources.

A 38-year-old man was admitted to the emergency room with a few days' history of severe pain on the right side of the neck. At the clinical examination, he was found to have an expanding right-sided neck mass. During the clinical examination, he experienced multiple episodes of hematemesis and suddenly died, despite medical intervention. Twenty-four hours after the death, a complete autopsy was performed. MPMCTA was carried out which showed a ruptured right common carotid artery, with contrast agent leaking through the rupture and into the mouth. The subsequent autopsy confirmed large hemorrhagic clots extending to the right side of the neck, pharynx, and larynx. An infusion of saline pumped by a syringe filled the right carotid artery showing a 3.5cm rupture of the left wall of the oropharynx. All the cervical organs were fixed in formalin solution for an accurate examination. The source of bleeding was identified as a 2cm tear in the wall of the right common carotid artery penetrating to the right side of the oropharynx through its previously described rupture. Abnormalities such as aneurysm, thickness, or thinness of the arterial wall were not detected. The etiopathology of the lesions was identified through histological examinations performed on vascular and oropharyngeal samples using Hematoxylin-Eosin (H&E) and immunohistochemical staining.

Spontaneous rupture of the common carotid artery is a very rare, life-threatening event. In the scientific literature, rupture is usually secondary to underlying pathology, principally aneurysmal disease, tumor invasion, prior heavy radiation exposure, or existence of infected oro-cutaneous fistula. In the presented case, the angiography can be useful to obtain the exact location of the site of bleeding.

In conclusion, CT-angiography is a useful tool to visualize the entire vascular system as part of the postmortem examination. MPMCTA can detect the exact source of hemorrhage in cases of sudden and unexpected death related to cardiovascular pathologies. Despite recent advances in diagnostic imaging, a complete methodological forensic approach by means of autopsy and histopathological examinations play an irreplaceable role in defining the exact cause of the death.

Postmortem CT-Angiography, Carotid Dissection, Sudden and Unexpected Death

G12 A Fatal Case of Pulmonary Tumor Embolism Due to Primary Cervical Carcinoma

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The goal of this presentation is to examine the histopathological aspects of late intracardiac metastases and of fatal pulmonary tumor embolism from previously removed carcinoma of the cervix, with diffuse and aggressive involvement of the right side of the heart and the pulmonary circulation.

This presentation will impact the forensic science community by describing the necessity of suspecting a myocardial metastasis in patients with an intracardiac mass and a history of uterine cervical cancer, with signs and symptoms of right cardiac failure and/or pulmonary embolism. According to scientific literature, early diagnosis is the only available strategy to improve prognosis in these cases.

Intracardiac metastases, although rare, can occur in patients with cancer. Manifestations include signs and symptoms due to intracardiac obstruction and low cardiac output or pulmonary embolism. Metastases may reach the heart via the lymphatic or hematogenous route or by direct or transvenous extension. Lymphatic spread tends to give rise to pericardial metastases, whereas hematogenous spread preferentially gives rise to myocardial metastases.¹ They may imitate valvular heart disease or cause cardiac failure, ventricular or supraventricular heart rhythm disturbances, conduction defects, syncope, embolism, or pericardial effusion. For carcinoma of the cervix, the most common sites of extrapelvic metastasis are the lungs, bones, or the cervical or supraclavicular lymph nodes. Cardiac metastasis is very rare with a frequency ranging from 1.6% to 8.0% and has a poor prognosis.²

This presentation reports a rare case of intracardiac metastases and fatal pulmonary tumor embolism caused by hematogenous metastases that mimicked pulmonary thromboembolism. A 54-year-old female had a history of uterine cervical carcinoma three years earlier, and was treated with radical surgery, chemotherapy, and radiation therapy. She was admitted to the hospital for persistent cough, hemoptysis, dyspnea, chest pain, and signs of right heart failure. A chest X-ray showed bronchopneumonia. A Computed Tomography (CT) scan of the chest demonstrated the presence of multiple pulmonary embolic phenomena involving the main pulmonary artery, the right lower and middle lobe arteries, and the left lower lobe artery with associated infarctions. Thrombotic material was also described within the heart. An echocardiogram showed an intracardiac mass which caused almost complete obstruction of the right ventricle from the tricuspid valve to the right pulmonary artery. During a right ventricular wall biopsy, the woman died and an autopsy was performed. The internal examination revealed the presence of a significant tumor mass occupying the entire right ventricle, infiltrating its free wall and the interventricular septum. The mass protruded into the pulmonary artery. Histological findings showed a poorly differentiated squamous cell carcinoma, compatible with primitive cell carcinoma of the cervix. Dissection of the pulmonary vessels revealed bilateral massive pulmonary embolism. The largest one measured 6cm in length and was histologically similar to the heart mass.

This case suggests that tumor embolism must be included in the differential diagnosis of respiratory symptoms in patients with a history of malignancy.

Pulmonary Tumor Embolism, Primary Cervical Carcinoma, Intracardiac Metastases

G13 Revelation of Paint Residue at the Skull by Stereomicroscopy Clarifies Criminal Circumstances

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After attending this presentation, attendees will better understand the benefits of bone examination by stereomicroscopy in cases of blunt violence against the skull. The detection of paint residues from the inflicting weapon can provide strong evidence of death circumstances which can easily be overlooked if diagnosis was only based on autopsy or a combination of autopsy and computed tomography.

This presentation will impact the forensic science community by improving the knowledge of how the stereomicroscopy, which is a simple and fast procedure, plays an important role in clarifying criminal circumstances.

Stereomicroscopy is a fast and simple diagnostic method. In this case, stereomicroscopy of a fractured area of the skull bone revealed otherwise invisible paint residue with a blue color. This finding was a crucial point in identification of the weapon which inflicted the skull fracture on the decedent.

A 56-year-old man was found dead in the stream of an urban area. The examination of the body at the autopsy revealed bruises, a laceration of the scalp, and an impression fracture on the right side of the frontal bone. The fracture was oval in shape, having a central elongated intrusion in the anterior direction, which was compatible with the impact of a blunt instrument. During the examination of the fracture area of the skull by low magnification stereomicroscopy, a very small fragment of blue paint was noticed. This finding led the police to investigate and look for a blunt object in the area. On the bottom of a lake, located close to the crime scene, a wallet belonging to the deceased, a knife, and a big blue pipe wrench were found. All items were lying close to each other above the sediment layer, suggesting that the items had been lying in the water for a relatively short amount of time.

Additionally, surveillance imagery from Closed-Circuit Television (CCTV) cameras, which had recorded the suspected perpetrator carrying a pipe wrench in his hand, were analyzed photogrammetrically. The analyses showed that the estimated length of the wrench, as calculated from the recordings, was in agreement with the length of the pipe wrench found in the lake. The form and size of the fracture intrusion were also compatible with the form and size of the pipe wrench found in the lake.

The piece of the skull bone with the paint residue was analyzed at the Danish Technological Institute. They found that the color of the paint on the skull matched the color of the paint covering the pipe wrench. The chemical structures of both binders and fillers in the paint residue were completely identical to the pipe wrench found in the lake. On the basis of this evidence and the perpetrator's own confession, he was found guilty by the court.

The presented case illustrates how careful examination of the fracture area by low-magnification stereomicroscopy may

provide strong evidence in terms of trauma and tool mark analyses. In this case, stereomicroscopy revealed otherwise invisible paint residue left by the pipe wrench, which led to identification of the weapon.

This study reveals that in cases of such complexity, the forensic pathological evaluation cannot be based only on the usual autopsy results, but should take into account the forensic technical evidence as well. A multidisciplinary team effort is absolutely preferable in the evaluation of such cases.

Stereomicroscopy, Multidisciplinary Effort, Homicide

G14 Bones Evidence in Dismemberment: A Patricide Case and a Review of the Literature

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After attending this presentation, attendees will be able to describe the impact of the forensic science in cases of dismemberment.

This presentation will impact the forensic science community by demonstrating the contribution of forensic science in solving cases in which the body is dismembered to answer questions about the weapon used and the cause and manner of death. In particular, the importance of the analysis of macroscopic bony margins with radiological and histopathological study regarding the vitality or otherwise of the injuries is stressed.

Introduction: This research explores the dismemberment that in reality is a relatively rare method of body disposal. After killing the victim, the murderer uses a very sharp cutting weapon (a saw, axe, etc.) to sever the limbs and cut the body into small pieces.¹ The term "dismemberment" describes the perpetrator's fragmenting the corpse or severing a part of a body in an act that is performed in bad faith.² The dismemberment of a body should not be confused with the signs of antemortem trauma. In many forensic dismemberment cases, it is difficult to discern the mode of death. Also, parricides are rare crimes, estimated to make up 1%-4% of all homicides and 20%-30% of homicides committed by psychotically ill individuals.³⁻⁵

Case Report: A case of patricide with dismemberment of the body by the daughter who suffered from schizophrenia is discussed. An elderly man had been missing for about one month. The police searched his apartment and found a number of boxes. Inside the boxes were many body segments, still covered by clothing with laces and ribbons, surrounded by lime and covered with cellophane. In this environment, there was no blood or insect larvae. When the victim's daughter was interviewed, she confessed only to the dismemberment, claiming to have found the body of her father already divided into two parts. The problems for investigators were to determine the vitality of the injuries inflicted on the body of the man, the time of death, and the manner in which the murder occurred. A judicial inspection was carried out and the postmortem computed tomography examination and autopsy with histopathological survey were performed. The soft tissues of the dismembered body segments were removed through maceration techniques and chemical treatment.

Conclusions: The analysis of bony evidence revealed

the presence of a “T”-shaped incision from a non-serrated cutting-edge instrument. The analysis of the bony margins determined the manner in which the victim was hit.

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Forensic Science, Dismemberment, Patricide

G15 Breed Designation for Unknown Equine Case Samples

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The goal of this presentation is to familiarize the animal forensic community with an existing cluster-based software that can be used to decipher breed admixture for unknown samples to obtain better match probabilities, not only in equine forensics, but also cases involved with breed-specific animals. This will also encourage researchers to work on a repository of breed-specific allelic frequencies that can be used for future forensic cases.

This presentation will impact the forensic science community by increasing awareness of the plethora of unsolved horse slaughter cases in the Miami-Dade and Broward counties of Florida which is a testimony to the gravity of and the increase in illegal slaughter activities. Often, with confiscated horsemeat, any physical description of the horse is lost and only DNA can link the horsemeat to evidence and to a suspect. Therefore, in order to more strongly correlate the evidence to the slaughtered horse, it is crucial to identify the breed of the horse as well as to obtain the individual equine profile. This calls for methods that will supplement individual identification with breed designation in the identification of unknown samples.

The current case study illustrates the use of population-genetic statistical software STRUCTURE 2.3.1 for breed identification of unknown equine samples of a slaughter case in Miami-Dade County, Miami, Florida. Allele frequencies of 15 domesticated breeds from published population studies were included in the analysis. Taking into account linkage and artifacts such as high stutter in some of the dinucleotide repeats for the 17 loci amplified, only 14 loci were included in the cluster analysis for determining the K populations. A K=8 was used to set the cluster analysis for correlated allele frequency as the 15 domesticated breeds showed similar allele frequencies. Prior to this cluster-based analysis, there was no substantial information that could

be used to determine breed-specific allele frequencies to aid in calculation of the Random Match Probability (RMP). Assuming the confiscated meat was from the more common local breeds found in South Florida, like Paso Fino, Quarterhorse, and Thoroughbred, the first analysis resulted in an RMP of 1.84×10^9 . By performing admixture analyses using all available breed allele frequencies, the case profile was found to be more closely aligned with the Standardbred, Lipizzaner, and Arabian breeds. Once the unknown horse's ancestry was established via the STRUCTURE analysis, using these published allele frequencies for genotype analyses, the RMP was recalculated and resulted in a better RMP of 1.63×10^{23} , 1.27×10^{20} , and 2.88×10^{23} , respectively.

This case study is an example for recommending the use of admixture software to determine the breed of unknown equine evidence in addition to the standard STR analyses as it can lead to a more relevant random match calculation.

Equine, Breed, STRUCTURE 2.3.1

G16 Severe Degenerative Tauopathy Following Closed Head Injury: Alzheimer's Disease vs. Chronic Traumatic Encephalopathy

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After attending this presentation, attendees will be able to identify the significance of traumatic brain injury on the development of spreading tauopathy and its potential as a pathogenic mediator of neurodegenerative disease.

This presentation will impact the forensic science community by highlighting the potential impact of a closed head injury in causing or accelerating the progression of chronic neurodegeneration, highlighting the role of tau protein as a toxic mediator of disease, and raises the issue of qualitative differences between chronic traumatic encephalopathy, aging, and Alzheimer's disease.

The role of head trauma in neurodegenerative disease has been recognized at least as far back as the 1920s. However, the role of head trauma in producing chronic disease has become a significant public health concern in recent years in light of the association between contact sports such as football, soccer, and boxing, as well as blast injuries in armed conflicts, with progressive degenerative tauopathy now termed chronic traumatic encephalopathy. History of closed head injury is also a known risk factor for Alzheimer's disease and confers an approximate twofold relative risk for development of disease. This presentation discusses the case of a 55-year-old woman who suffered a fall at a department store resulting in multiple cerebral contusions. Following the fall and the closed head injury, she developed frequent seizures and increasing confusion. She expired three years after the closed head injury. At autopsy, she had remote contusions with encephalomalacia involving the orbitofrontal regions bilaterally, the bilateral temporal poles, and the right posterior medial temporal lobe. Hydrocephalus *ex vacuo* and a cavum septum pellucidum were also present. Microscopic examination revealed extensive neurofibrillary degeneration using phospho-tau (AT8) immunohistochemistry, qualifying for advanced/end-stage Alzheimer's disease (Braak stage VI). Extensive amyloid plaques throughout the neocortex were also noted on histology. The pattern of phospho-tau deposition overlapped between that seen in chronic

traumatic encephalopathy (superficial layer, perivascular, glial tau), and the pattern seen in Alzheimer's disease. This case highlights a potential role of head trauma in accelerating neurodegeneration and raises the issue of spreading tauopathy as a pathogenic mediator of neurodegenerative disease.

Traumatic Encephalopathy, Tauopathy, Alzheimer's Disease

G17 Video Surveillance: A Further Useful Aid in Crime Scene Investigations and Crime Reconstruction

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After attending this presentation, attendees will understand that in Crime Scene Investigations (CSI), the requirement of multidisciplinary competences is fundamentally important for the analysis, collection, and interpretation of the evidence next to a corpse and the possibility of using recordings of common video surveillance cameras to provide invaluable information to verify and confirm the hypotheses based on objective findings.

This presentation will impact the forensic science community by showing four original cases in which the analysis of video recordings by surveillance have helped medicolegal evaluations and event reconstructions and thus enabled the elimination of many doubts about the course of events.

Crime is not only associated with particular implementation arrangements but also with testimonial data that is absent or potentially controversial. In these cases, laboratory data are not always sufficient to achieve reliable conjectures and can even fail to reach the limits of "beyond any reasonable doubt." In such cases, inappropriate acquittals or convictions can occur. In this context, the video recordings of surveillance cameras can and often do help forensic assessments and reconstruction of events resulting in the correct resolution of cases.

The first case involves a robbery attempt at a gas station where the manager fired a pistol at two robbers fleeing on a fast motorcycle; this resulted in the "accidental" death of one of the robbers. It was thought that the bullet had hit the victim after it ricocheted off the ground. Examination of video surveillance recordings showed that the angle of the motorcycle could only mean a seemingly upward trajectory of the bullet. It was found that the bullet crossed the seat but was undeformed before striking the passenger on the motorcycle, so the possibility of a ricochet was eliminated.

In the second case, two masked gunmen broke into an arcade where about 20 customers were playing games. Four shots were fired, one of which killed a young man. According to eyewitnesses, the victim was lying on the ground with his back to the wall of the room, partially sheltered behind a table and some chairs. The inspection of the area was compared with the records of a video surveillance camera which allowed for the reconstruction of the moment when the armed robber pointed the gun downward and toward where the victim was located. This "frozen" position coincided perfectly with the reconstruction of the trajectory of the bullet and demonstrated that there had been an impact and ricochet from the floor. This hypothesis was also supported by the particular deformation of the projectile recovered at the autopsy and with its trajectory in the victim's body.

The third case involves a young African male who was found dead in the swimming pool of a private home where he

worked as a gardener. During the investigation, it was suspected that the victim, who could not swim, had been thrown into the water during an altercation with some friends. The autopsy showed there were no signs of a struggle and the cause of death was in fact drowning. The records of a video surveillance camera showed that the victim had intentionally jumped into the deep end of the pool and had made no attempt to save himself. The case was dismissed as a suicide.

In the fourth case, a young Armenian male died in a hospital from severe head injuries. Some friends reported that he had accidentally fallen from the top of a wall but the trauma found at the autopsy were compatible with injuries sustained from kicks and punches to the head. The recordings of a video surveillance camera showed that there had been a dispute between some people and that the victim had been repeatedly kicked and punched in the head.

This presentation illustrates that the competencies of the medical examiner and law enforcement officers can lead to results that can be supported by images from video surveillance cameras. These images can reveal what actually transpired and thus provide the court with irrefutable evidence to solve a crime.

Video Surveillance Devices, CSI, Crime Reconstruction

G18 It Must Be Suicide! Methodological Approach in Atypical Hanging With Handcuffs

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The goal of this presentation is to show the difficulties in distinguishing between suicide or homicide when an atypical hanging occurs and how to collect undisguisable features.

This presentation will impact the forensic science community by demonstrating the relevance of a complete methodological approach in all cases of hanging in which suicide is doubtful. A complete crime scene investigation has to be followed by autopsy and lab tests (histology, immunohistochemistry, toxicology). Radiological investigations should also be performed.

Hanging is one of the most commonly used methods for suicide worldwide. Distinguishing a potential homicidal hanging from a suicide poses difficult challenges for forensic pathologists. A variety of staged crime scenes are encountered by investigating officers. A suicide may be staged as a homicide by family members or friends in an attempt to avoid embarrassment or for other personal, social, or religious reasons. Staging a suicide as a homicide may be a final effort by the victim to gain popularity, attention, take revenge against friends or family, or to ensure his killing. In contrast, some suicidal crime scenes could be highly suspicious for homicide. A case of atypical hanging is presented. A 52-year-old man was found lifeless in his house hanging from a rope wrapped around his neck and anchored on a pipe in the ceiling. His wrists were bound by a pair of handcuffs. His arms were extended and adducted to the abdominal wall. His trousers were down to his knees. His clinical history was remarkable for depressive syndrome with several unsuccessful suicide attempts. The prosecutor's office alerted the forensic pathologist and a detailed crime scene investigation was performed. House inspection was unremarkable and no suicide note was found. The body temperature was 31°C, rigor mortis was present, and fixed hypostasis was observed in the upper and lower limbs. A ligature

mark encircling the neck with bruises was described at external examination. Red-bluish areas were found on the wrists after the handcuffs were removed. Subconjunctival hemorrhagic petechiae were also recorded. Traumatic injuries were excluded after external examination and Computed Tomography (CT) scan investigation. A complete postmortem examination was performed the next day. Mild cerebral and pulmonary edema with white foam in the main bronchi were detected. The right sternocleidomastoid muscle had a mild focal hemorrhage. Pulmonary edema, congestion, and focal pulmonary atelectasis were identified by microscopic analysis of routine Hematoxylin-Eosin (H&E) stains. The microscopic observation of the skin sections from the ligature marks revealed intra-epidermal mild leukocyte reactions and alteration of the musculature in the form of Zenker's necrosis. In addition, immunohistochemical staining of skin samples was performed utilizing antibodies to antitrypsin, fibronectin, TNF α , IL-6, IL-8, IL-10, MCP-1, IL-15, IL-1 β , CD45, CD4, CD3, CD8, CD68, CD20, and CD15 to confirm vitality of reactions in the ligature mark. Toxicological analysis on blood and urine were unremarkable. Mechanical asphyxia by suicidal hanging was indicated as the cause of death.

Binding the wrists to prevent rescue attempts is rare in suicidal hanging and is rarely reported in forensic literature. These cases represent a difficult challenge in distinguishing between homicide or suicide. A thorough postmortem investigation will allow the pathologist to make the correct determination.

Atypical Hanging, Handcuffs, Suicide-Homicide

G19 Globus Pallidus Necrosis Unrelated to Carbon Monoxide Poisoning: Retrospective Analysis of 24 Cases of Basal Ganglia Necrosis

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After attending this presentation, attendees will understand that, in contrast to textbook descriptions of globus pallidus necrosis, this lesion is rarely associated with and certainly not specific for Carbon Monoxide (CO) poisoning.

This presentation will impact the forensic science community by critically examining the assertion that globus pallidus necrosis is a typical finding of CO poisoning and will contribute to the understanding of causes and mechanisms of globus pallidus injury.

It is well demonstrated that CO poisoning can cause central nervous system injury in a variety of locations, including the basal ganglia, cerebral cortex, hippocampus, and cerebellum, with delayed CO exposure more associated with globus pallidus, basal ganglia, and white matter injury (e.g., Grinker's myelinopathy). Globus pallidus necrosis, especially when seen bilaterally, is described in forensic textbooks as a lesion characteristic of, or classically seen in, delayed CO poisoning. However, this description conflicts with a number of published reports. A literature search revealed that globus pallidus necrosis has been observed in a wide variety of cases, including different types of drug overdose, post-anesthesia, in children after cardiac surgery, prolonged coma, cerebral arteriosclerosis, hemolytic uremic syndrome, acute renal failure, metabolic disorders, viral infections, and wasp stings.

To test the hypothesis that globus pallidus necrosis is characteristic of CO poisoning, this study examined autopsy cases from the King County Medical Examiner's Office between 1994 and 2013 in which globus pallidus or basal ganglia necrosis was

photographically documented. There were a total of 39 cases, in which 15 showed unilateral hemorrhagic lesions of the basal ganglia which were subsequently eliminated from analysis, since they were either due to hypertensive stroke or trauma and since none were related to carbon monoxide exposure. This left 24 cases with necrotic or cystic lesions of the globus pallidus or basal ganglia, 14 of which had bilateral lesions. For comparison, two cases of delayed CO poisoning were identified.

The 24 cases of necrotic basal ganglia lesions were examined for the associated or causative disease or injury with the following results: nine were drug-related deaths or had significant drug abuse history; six were due to hypertensive and atherosclerotic cardiovascular disease; two were due to Huntington's-like disorders; and two were due to asphyxia (one infant with positional asphyxia and one adult with compressional asphyxia). Of the remaining five cases, there was one case of each of the following: remote trauma; delayed drowning; rheumatic heart disease; chronic ethanolism; and cerebral artery gas embolism in a scuba diver. Seven of the 24 cases had a clinical diagnosis of anoxic encephalopathy. No cases were identified with basal ganglia or globus pallidus necrosis associated with CO poisoning, and the single case dying after prolonged coma following CO monoxide poisoning had no evidence, gross or microscopically, of globus pallidus or basal ganglia necrosis.

The results of this study indicate that globus pallidus and basal ganglia necrosis is a consequence of cerebral hypoxia from a variety of causes and is not specific for any type of injury. However, selective hypoxic injury of the basal ganglia is not a common feature of global cerebral hypoxia and is likely due to factors of cerebral blood flow that are not well understood. Finally, this study provides no support for considering globus pallidus necrosis to be characteristic of CO poisoning.

Carbon Monoxide Poisoning, Cerebral Hypoxia, Globus Pallidus Necrosis

G20 Examination of the Cervical Spinal Cord in Infants and Toddlers

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After attending this presentation, attendees will understand the methodology and different techniques available for examination of the cervical spinal cord and adjacent structures in infants and toddlers to properly diagnose trauma, malformations, or natural disease processes resulting in death.

This presentation will impact the forensic science community by demonstrating the advantages of the *en bloc* examination of the neck structures, including the spinal cord, vertebrae, neurovascular, and adjacent soft tissues over the standard post-extraction spinal cord examination in death investigations of infants and toddlers.

Abnormalities of the spinal cord and surrounding tissues, due to malformations or other processes whether traumatic, ischemic, or vascular, could either explain death or develop after the

event that ultimately led to death. The differentiation between these two possibilities is the key to correct diagnosis and interpretation. Although CT scan is considered the best imaging method to rule out cervical skeletal abnormalities or injuries, it lacks sensitivity to diagnose soft tissue pathology. Injuries to spinal cord parenchyma, such as bleeding, may be identified on CT imaging when large enough, but due to the small size and naturally and technically occurring artifacts, evaluation of the parenchyma, nerves, and ganglia is limited. Furthermore, in contrast to the cerebral region, radiographic separation of subarachnoid, subdural, and epidural spaces in the cervical region by CT is very difficult. The standard assessment of the cervical spinal cord including cervicomedullary junction is challenging due to visualization and removal difficulties and the possibility of adding artifact during the removal process. Maintaining the anatomic relation of the spinal cord, nerve roots, and vascular supply with the surrounding osteocartilaginous structures provides more precise assessment of the individual elements with connections and surrounding spaces that aid in the pathophysiologic interpretation of the findings. Specifically, if the subarachnoid, subdural, and epidural spaces remain in place, the intraspinal vertebral artery as well as the points of entry and exit can be examined and the nerve roots and root ganglia can be followed.

Cases included non-sequential infants and young children who had an autopsy at the Office of the Chief Medical Examiner (OCME) in Baltimore, Maryland. The cervical spines and surrounding tissues were removed *en bloc* from the occipital base to the upper thoracic spine following previously described dissection techniques. After decalcification and horizontal serial sectioning, the tissue blocks were processed and whole mount sections at all levels were obtained and stained with Hematoxylin-Eosin (H&E) and additional special stains, if needed, included immunohistochemical stain with beta-Amyloid Protein (APP). Postmortem CT scans obtained at the OCME were blindly evaluated by two separate radiologists and a forensic pathologist with radiologic training. The results of the CT and pathologic evaluation of the neck in a group of children less than four years of age will be presented.

Cervical Cord, Infants and Toddlers, CT and Histologic Examination

G21 Histopathological Examination of Childhood Optic Nerve Sheath Hemorrhage

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The goals of this presentation are to identify the vasculature associated with the optic nerve sheath, discuss possible mechanisms of optic nerve sheath hemorrhage, and identify the difference between an intradural, a subdural, and a subarachnoid hemorrhage.

This presentation will impact the forensic science community by increasing awareness of the similar hemorrhage patterns of optic nerve sheath hemorrhage associated with fatal intracranial bleeding in young children from natural and traumatic causes.

Although Optic Nerve Sheath Hemorrhage (ONSH) has long been recognized as a complication of intracranial bleeding and sudden increased intracranial hypertension, the vascular source of optic nerve sheath intradural, subdural, or subarachnoid hemorrhage is unknown. Muller and Deck noted ONSH in 87% of 46 eyes examined after a sudden rise in intracranial pressure, while only 37% of the eyes had retinal hemorrhages.¹ Walsh and Hedges demonstrated that ONSH was not a direct extension

from intracranial bleeding to the optic nerve and retina, but these hemorrhages occur secondarily to sudden distension of the optic nerve subarachnoid space from an acute rise in intracranial pressure, which presumably ruptures the bridging vessels in the optic nerve sheaths.²

Lambert *et al.* first described the presence of ONSH in Shaken Baby Syndrome (Abusive Head Trauma).³ Subdural ONSH has since been observed in 65% to 100% of infants and young children diagnosed with Abusive Head Trauma (AHT). The most common site for AHT associated ONSH is in the immediate retrobulbar portion of the optic nerve similar to the findings in non-abusive intracranial bleeding observed by Muller and Deck and Walsh and Hedges. Lambert *et al.* suggested that an acute rise in intracranial pressure, transmitted to the optic nerve sheath, caused expansion of the sheath with rupturing of bridging veins. However, Wygnanski-Jaffe *et al.* suggest that orbital and optic nerve injury is more common and more severe in AHT than accidental head trauma.⁴ They regard the eye and its orbit as a single unit, damaged directly by the mechanical process of shaking and surmise that there is a unique feature of repeated acceleration-deceleration injury, as seen in SBS, which leads to orbital trauma with retinal hemorrhages and ONSH. They suggest the increased frequency of hemorrhage at the anterior and posterior optic nerve sheath indicate that the junction of the globe and optic nerve and the firm posterior orbital attachments of all orbital structures create fulcrums leading to tissue damage at these locations.

Although optic nerve bridging veins have been implicated as the bleeding source of ONSH, this has not been confirmed histopathologically and does not adequately explain the increased concentration of intra-dural hemorrhage that is typically seen to communicate with subdural blood near the retro-bulbar optic nerve sheath. Returning blood from the retina and optic nerve, the central retinal vein may remain for some distance in the substance of the dural sheath before joining the superior or sometimes the inferior ophthalmic vein, or even the cavernous sinus directly.

This study hypothesized that serially sectioning and examining the retrobulbar optic nerve and surrounding sheath histologically should identify disrupted blood vessels within the intradural portion of the ONSH and thus explain the vascular source of ONSH associated with intracranial bleeding in children. Six archived autopsy cases with bilateral ONSH in children with fatal intracranial injuries were retrieved: two traumatic brain injuries from child abuse; two accidental traumatic brain injuries; and, two spontaneous non-traumatic intracranial hemorrhages.

Their ages were from 2 to 55.3 months and the male:female ratio was 1:1. Survival time from injury or incident to death varied from 2.75 to 57 hours. All had intra-cranial hemorrhage and bilateral retinal hemorrhages.

The optic nerves and surrounding sheaths were sequentially sectioned at 4 microns (thickness) with alternating Hematoxylin-Eosin (H&E) stained and unstained sections (50 sections/optic nerve: (25 H&E slides and 25 unstained slides)). All H&E histological sections were examined and assessed for intra-dural hemorrhage, subdural hemorrhage, subarachnoid hemorrhage, and perineural soft tissue hemorrhage.

A similar pattern of hemorrhage was identified in all six cases. There was no significant variation in the location of the extravasated blood in the natural, accidental, or child abuse deaths. All cases exhibited asymmetrical intradural hemorrhage that was centered on intra-dural blood vessels and all had perineural soft tissue hemorrhage. This study was unable to identify a definite disruption of the retinal artery, retinal vein, and/or accompanying smaller vasculature with the number of sections examined. A similar pattern in these cases suggests a common source of the intra-dural, subdural, and subarachnoid blood.

To identify the vascular source of the ONSH blood, additional serial sectioning must include the entire retrobulbar

optic nerve and sheath including the entire intra-dural course of the retinal artery and vein. Careful orientation of the optic nerve segments will also facilitate assessment. Finally, application of an elastic-trichrome stain on alternating sections will assist in illustrating the intra-dural vascular walls that can be obscured by the extravasated blood.

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Forensic Science, Optic Nerve Sheath Hemorrhage, Retinal Hemorrhage

G22 Ethylene Glycol Toxicity: Death Before Calcium Oxalate Crystal Deposition

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After attending this presentation, attendees will recognize that Ethylene Glycol (EG) can result in fatal toxicity in the absence of the pathologic clue of crystal deposition in the kidneys or brain.

This presentation will impact the forensic science community by raising the threshold of suspicion of EG toxicity in the absence of birefringent crystals in the kidney, which is currently the "gold standard" marker that triggers chemical analysis of EG.

EG is a key ingredient in automobile antifreeze. It is an odorless, colorless, sweet-tasting liquid that causes intoxication and death if consumed in sufficient amounts. In attempts at suicidal overdoses, its toxicity is expressed in three clinical phases of poisoning according to post-ingestion time. Much like ethanol, the first stage is characterized by direct Central Nervous System (CNS) depression, which occurs shortly after ingestion and lasts for several hours. This period involves drowsiness, disorientation, and confusion; affected individuals may appear drunk. Convulsion, stupor, and coma may develop in the next stage, about 12-24h after ingestion. In this stage, EG metabolites cause severe non-ketotic metabolic acidosis with an elevated anion gap, cardiopulmonary manifestations, and possible multisystem organ failure. In many cases of EG poisoning, the degree of acidosis can be a prognostic factor, but CNS manifestations are also potentially fatal. In the third stage, about 24-72h after ingestion, a well-known pathological feature is the formation of microscopically visible calcium oxalate crystals from the metabolism of EG into oxalic acid and calcium chelated precipitates. It takes approximately 24 hours for signs of renal impairment to occur and for calcium oxalate crystals to appear in the urine.

To date, no confirmed fatal cases of EG toxicity without tissue birefringent crystals have been reported in the literature. EG is not able to be detected by a volatile panel or traditional toxicology screens; as such, analysis typically depends on suspicion and a special request.

Presented are two cases of fatal EG ingestion in which

the characteristic birefringent crystals were absent. Case 1 is a 56-year-old White male found in a field with a note on him, suggestive of a suicide note. He had prior expressed suicidal ideation with stated intent to ingest EG. Because of the history of recent suicidal ideation with specific intent, an EG analysis was requested. EG was overwhelmingly positive: 23,296mg/L in urine, 7,974mg/L in peripheral blood, 12,446mg/L in vitreous humor, and 52,175mg/L in stomach contents. No birefringent crystals were seen in the kidney or brain. The cause of death was certified as EG toxicity and the manner of death was suicide.

Case 2 is a 53-year-old White female found in her residence with a suicide note. Her past medical history included bipolar disorder and prior suicide attempts in which she ingested EG, ethanol, acetaminophen, and paroxetine. Autopsy findings were significant for severe stenotic atherosclerosis of the left anterior descending branch of the left coronary artery. Initial toxicology did not detect ethanol or acetaminophen. Paroxetine was within a therapeutic range. The kidney and brain were negative for birefringent crystals. Further analysis for EG was requested because of the history. EG was in excess of 1,113mg/L in urine and vitreous humor, and 937mg/L in heart blood. The cause and manner of death were classified as EG toxicity and suicide, respectively.

Death in massive EG ingestion may be similar to acute ethanol toxicity. Like ethanol, EG has a low volume of distribution, so it is rapidly absorbed with peak concentrations occurring 30-60 minutes after ingestion. EG also increases gamma-aminobutyric acid receptor activity in the CNS, which causes sedative effects including severe confusion, unconsciousness, and respiratory depression creating a respiratory acidosis that precedes the metabolic acidosis caused by accumulation of glycolic acid. Therefore, EG can exert profound toxicity even before its transformation into more toxic metabolites such as glycolate and the hallmark deposition of calcium oxalate crystals.

It is proposed that EG ingestion in sufficient quantities may precipitate death before crystal deposition in the tissues. This study recommends analysis for EG in all cases with a history suspicious for EG ingestion — or in cases with a history suspicious for suicide with negative initial toxicology testing — even in the absence of birefringent crystals.

Ethylene Glycol, Postmortem, Toxicology

G23 Heroin Overdose — Just Another Reason to Lose Those Extra Pounds?

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After attending this presentation, attendees will learn about the correlation between Body Mass Index (BMI) and postmortem morphine levels in heroin mono-intoxication deaths.

This presentation will impact the forensic science community by providing a comprehensive analysis of heroin mono-intoxication deaths with BMI.

Background: Overweight (OW) and Obesity (OB) are the most common nutritional disorders, with 69% of the United States population overweight or obese and 28% of Maryland's population obese. Obesity is linked to a wide range of chronic and acute disease, and previous studies have shown a strong

association between obesity and premature death. BMI is an excellent parameter to assess OW and OB. A BMI greater than 30 predicts a 50%-100% increase in premature death from all causes as compared to a normal BMI of 18-25. Most of these deaths are the result of cardiovascular disease. This study hypothesized that the OW/OB population would be at higher risk of sudden death at lower morphine levels during heroin intoxication due to respiratory dysfunction and Sleep Apnea/Obesity Hypoventilation Syndrome (SA/OHS). This hypothesis was evaluated via a retrospective study of the correlation between BMI and postmortem morphine levels in cases of fatal heroin mono-intoxication deaths.

Materials and Methods: A 10-year retrospective review of death certificates between 2003 and 2012 was performed at the State of Maryland Office of the Chief Medical Examiner (OCME) for deaths certified as "heroin" or "narcotic (heroin)" intoxication or overdose. All causes of death were confirmed by case history, scene findings, and 6-monoacetylmorphine positivity. Causes of death due to a mixed drug intoxication involving heroin and other drugs or alcohol were excluded. Also excluded were cases in which "cocaine use" was considered as contributing to death. The BMI was calculated and classified as normal (18-24.9), overweight (25.0-29.9), obese class I (OB 1, 30-34.5), and obese class II (OB 2, >35.0). Postmortem blood morphine levels were determined in either peripheral or heart blood based on levels as measured in the OCME toxicology lab. The deaths were further analyzed based on postmortem blood morphine ranges of: <50, 50-400, and >400ng/ml. Median morphine levels in various subgroups were calculated. Median rather than mean morphine level was used because in certain cases the toxicology lab reported morphine levels as ">500" or ">1,000ng/ml"; also, median is a more robust measure of central tendency and thus less influenced by outliers.

Results: A total of 398 heroin mono-intoxication deaths were identified. Of these subjects, 29% (116) had a normal BMI, 38% (152) were OW, 22% (89) were OB 1, and 10% (41) were OB 2. The median postmortem morphine (ng/ml) for each BMI group was as follows: normal: 135; overweight: 170; OB 1: 160; and OB 2: 150. However, there was significant scatter of morphine levels at all levels of BMI and no clearly statistically significant trends were identified. Subgroup analysis restricted to deaths with postmortem morphine levels > 400 ng/ml revealed the following incidences: normal: 11.2 % (13); overweight: 17.8% (27); OB 1: 15.7% (14); and OB 2: 9.8% (4).

Discussion: Being overweight or obese is generally thought to be an unhealthy condition with attendant comorbidities that increase the risk for premature death. Research results showed greater than 70% of all heroin mono-intoxication deaths were overweight or obese individuals, which was slightly higher than the representation of these individuals in the general U.S. population. This study showed 33% of the cases were obese, which is higher than the percentage of obese in the state of Maryland (28%). An over-representation of obese individuals dying of heroin overdose may reflect a basic susceptibility of these individuals to dying during heroin abuse, increased heroin use by these individuals, or that the OCME population is skewed. However, the median levels of postmortem blood morphine were similar in all BMI groups. An interesting trend in subgroup analysis was that the OB 2 group had the lowest incidence of high postmortem morphine blood levels. Several possibilities for this trend exist, such as different routes of administration of morphine in morbidly obese individuals (e.g., lower rates or percutaneous injection due to the difficulty of finding veins; and possibly different social settings of drug use in morbidly obese people).

Heroin, Obesity, Body Mass Index

G24 Bicyclist Fatalities in New York City

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After attending this presentation, attendees will understand the epidemiology, risk factors, and types of injuries in bicycling fatalities observed in the New York City area.

This presentation will impact the forensic science community by promoting understanding about fatal bicycling injury patterns and the risk factors for these fatalities in a large metropolitan area.

According to the Department of Transportation, New York City has over 250 miles of designated bicycle lanes and over half a million residents use a bicycle at least several times a month. Bicyclists share the roadway with motorists including buses and trucks. The medical examiner case files were reviewed on all bicycling fatalities in New York City to study the autopsy findings, toxicological results, and the epidemiological patterns of these fatalities in a large metropolitan area.

The New York City Office of Chief Medical Examiner electronic death certificate database was searched for all fatalities involving a bicycle that occurred between April 2006 and August 2012. There were a total of 156 fatalities (ten were excluded from the study: five pedestrians struck by a bicycle; one adult rode a child's tricycle; one fell onto a bicycle; and three accidents occurred outside New York City). The following data were collected: age; sex and ethnicity of the bicyclist; reason for cycling and helmet use; survival interval; New York City borough; time and place; environmental conditions; type of bicycle and motor vehicle; autopsy findings (injury locations and skeletal fracture sites); and, toxicological results.

Of the 146 bicyclists, there were 134 males and 12 females and the average age was 40 years (range 8-83 years of age). The ethnicities were White (n=48), Hispanic (n=43), Black (n=32), Asian (n=19), and unknown (n=4). Recreational use accounted for 40 fatalities and work commuting for one. There were 5 work-related deaths, and 100 were unspecified. A survival interval occurred in 72 bicyclists (from 2 hours to 33 years). Within the five boroughs of New York City, the fatalities were distributed as: Brooklyn=52; Manhattan=42; Queens=33; Bronx=19; and Staten Island=0. Among those that were known, the most frequent time when the fatalities occurred was between 6:00 p.m. and 7:00 p.m. (n=11) and 10:00 p.m. and 11:00 p.m. (n=11), with 62% (n=86) occurring between 1:00 p.m. and midnight. Most fatalities occurred on a Friday (n=28). The environmental conditions reported in 80 instances were: clear (n=55); raining (n=7); cloudy (n=6); cold (n=2); or hot (n=1). Most fatalities occurred during the summer months (n=62) with 24 of these occurring in the month of August. The lowest number of fatalities was recorded during winter months (n=19). The most frequent motor vehicle involved was a four-door sedan (n=34) or truck (n=18). The type of bike involved was recorded in only 11 instances (4 were mountain bikes). For helmet use, 15 decedents were wearing a bike helmet and 67 were not (64 cases did not document this information).

Postmortem examination included 116 autopsies and 30 external-only examinations. All causes of death were due to blunt trauma and all manners of death were accidents. The most commonly injured body region was the head (n=121) and 91% of fatalities who did not wear a helmet sustained a head injury. Other regions included: thorax (n=72); abdomen and pelvis (n=54); and upper/lower extremities (n=51). Vertebral injuries occurred in 37 instances (most commonly the cervical spine in isolation (n=17) of which 82% had an associated head injury). Fractures, detected

in 132 fatalities, most commonly involved the skull (n=89) and/or ribs (n=64). Toxicological analysis detected ethanol (n=22) with an average blood concentration of 0.13gm%, cannabinoids (n=15), cocaine (n=11), and methadone (n=3).

Similar to fatalities in motor-vehicle-only collisions, bicyclists also die from blunt injuries, and drug/ethanol intoxication is not unusual. Although some case information may not be available in some deaths (especially if there is a prolonged survival interval), investigators need to concentrate on obtaining information on the use of bicycle helmets, reason for bicycling, and type of bike (e.g., wide handle bars, which may increase the likelihood of a collision). The overwhelming male predominance may be a reflection of a higher number of male bicyclists and/or related to increased risk taking among males. This information may be useful for public health considerations when looking for ways to prevent these deaths.

Fatality, Bicycling, Trauma

G25 Evaluation of Myocardium Damage at the Right Ventricle Compared to the Left One — Improvement of a Diagnostic Tool for the Diagnosis of Fatal Pulmonary Fat Embolism?

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After attending this presentation, attendees will improve their knowledge of the physiopathology and pathology of cardiac damage after pulmonary fat embolism. They will learn how immunohistochemistry can be helpful in the diagnosis of fresh ischemic cardiac changes and how to interpret right ventricular ischemia in cases of pulmonary fat embolism.

This presentation will impact the forensic science community by providing useful tools for better understanding the phenomenon of fatal pulmonary fat embolism and its cardiac effects.

Fat Embolism (FE) is a common complication of blunt force injuries occurring in major trauma, especially if fractures of long bones are present. In cases of fulminating FE, the sudden massive obstruction of the pulmonary circulation results in rapid and often lethal increase in the impedance to right ventricular ejection with subsequent right heart ischemia and failure.

A method to evaluate the occurrence of right ventricular ischemia resulting in acute right heart failure in cases of severe pulmonary FE has been proposed. This method allows for the morphological diagnosis of primary right heart failure due to acute persistent pulmonary obstruction. The major limits of this work are bound to its retrospective character in that immunohistochemical analyses were performed on relatively few cases of pulmonary FE on available paraffin-embedded blocks of cardiac tissue, collected at autopsy on the basis of a routine sampling protocol without an extensive systematic investigation of different anatomical regions of the heart. The study presented here has different goals. First, it proposed to validate the preliminary study by investigating a more consistent number of cases included in a prospective protocol. Furthermore, there was interest in studying whether right ventricular damage is homogeneously distributed in the different regions of the right ventricle in cases of severe fatal FE. Finally, the question of

the role of this method as a potential tool in the improvement of the medicolegal diagnosis of fatal pulmonary embolism was addressed during this study.

In a prospective study, 220 consecutive autopsy cases performed at the University Center of Legal Medicine in Geneva, Switzerland between July 2010 and March 2012 were investigated. In each case, eight cardiac regions (anterior, lateral and posterior wall of the right and the left ventricle, and anterior and posterior part of the interventricular septum) were sampled and standard histology staining (Hematoxylin-Eosin, Masson's trichrome) was performed. Immunohistochemical reactions with the antibodies against Fibronectin and the terminal complement complex C5b-9 were performed. FE was determined by means of frozen sections of the lungs (one sample from each lobe was collected and investigated), the central nervous system (one sample from the cerebral and cerebellar cortex and from the pituitary gland was collected and investigated), and the kidneys (one sample from each kidney). The frozen sections were stained with oil red O.

The slides were investigated by two different observers with final consensual evaluation. In cases of discord, a third forensic pathologist gave his advice and allowed final decision. Classical histology signs of fresh cardiac damage such as hypereosinophilia, presence of contraction bands, myocytolysis, fragmentation of the cardiomyocytes, interstitial hemorrhage, and inflammatory infiltrates were systematically searched and classified into four degree categories: absent, weak, moderate, and severe. Similarly, the immunohistochemical reactions against the antibodies Fibronectin and C5b-9 were classified into four degree categories: negative reaction, single cell reaction, group cell reaction, and diffuse reaction. The degree of FE was determined following the method proposed by Falzi.

In this presentation, the results of this study will be presented and the implications for routine medicolegal investigation of FE will be discussed.

Fat Embolism, Right Ventricular Damage, Ischemia

G26 Commode Cardia — Death by Valsalva Maneuver: A Case Series

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The goal of this presentation is to report the incidence of decedents who died on the toilet following the Valsalva maneuver during the process of defecation (also known as "Commode Cardia") in order to better understand the impact and importance of this phenomenon.

This presentation will impact the forensic science community by reviewing the possible pathophysiology of Commode Cardia, providing information regarding the incidence of Valsalva maneuver and cardiovascular disease, and stratifying the importance of the Valsalva maneuver as a minor risk factor for sudden cardiac death in the presence of underlying arteriosclerotic cardiovascular disease.

It is well documented that vagal stimulation causes cardiovascular changes through autonomic stimulation. Various techniques that induce vagal stimulation have been identified and are called vagal maneuvers. In the clinical medical setting, these maneuvers are routinely applied for therapeutic or diagnostic reasons to utilize known specific cardiovascular changes. However,

some of these vagal maneuvers may be involuntarily used during normal daily activity and lead to unintended and severe cardiac consequences. One example of this is the Valsalva maneuver during the act of defecation.

The Valsalva maneuver is divided into four phases. In phases I and II, there is increased intrathoracic pressure that reduces the amount of blood flowing into the thoracic cavity, especially in the vena cava, aorta, and cardiac chambers. A small but significant decrease in the coronary flow velocity also occurs in phase I. For these reasons, the clinical application of the Valsalva maneuver may be contraindicated in a variety of cardiovascular conditions, including hypertrophic obstructive cardiomyopathy, significant aortic valvular disease, recent myocardial infarction, and aortic stenosis. However, individuals with these cardiovascular conditions who involuntarily Valsalva during evacuation of the colon could potentially suffer adverse events such as a syncopal episode through decreased cerebral blood flow and cerebral perfusion, sinus pause, atrioventricular block, arrhythmia, or myocardial infarct from decreased flow through coronary arteries. Thus, the Valsalva maneuver can be potentially fatal, a chain of events that can be described as "Commode Cardia." Although this phenomenon has been well documented, little is known about the incidence of this particular pathophysiologic event.

In 2012, the Wayne County Medical Examiner's Office in Detroit, Michigan, investigated 127 deaths located in the bathroom. Twenty people died on the toilet: 13 of cardiac disease following a Valsalva maneuver (natural); 4 from drug intoxication (accident); 1 from incised wounds to the wrist (suicide); 1 from a gunshot wound to the head (suicide); and 1 from smoke and soot inhalation (accident). In another 32 cases, the possibility that the decedent used the toilet just prior to having the fatal cardiac event could not be ruled out. During that same period, the office signed out a total of 608 cases of hypertensive and arteriosclerotic cardiovascular disease and arteriosclerotic cardiovascular disease. Based on this data, the incidence of Valsalva maneuver immediately preceding a terminal cardiac event, or Commode Cardia, is between 2.14% and 7.4% in our patient population. This study clearly indicates that the Valsalva maneuver is a small but significant risk factor for sudden cardiac death in the presence of underlying arteriosclerotic cardiovascular disease.

Commode Cardia, Valsalva Maneuver, Cardiovascular Disease

G27 Human Fatalities Due to Blunt or Penetrating Animal Injuries

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After attending this presentation, attendees will better understand the pattern of animal-related injuries in human victims.

This presentation will impact the forensic science community by providing evidence in human casualties due to animal attacks in Turkey.

Animal-related injuries in Turkey are not well documented

in the world scientific community. People engaged in animal husbandry or agriculture in rural areas are at increased risk for fatal and non-fatal injuries. This study uses the following six cases to aid the scientific community of Turkey in establishing epidemiological data for these types of injuries.

Case 1: An 85-year-old male with penetrating injuries to the lumber area was injured by the horn of a runaway cow which escaped from being slaughtered. The man died in the hospital on the same day. After the necropsy, the cause of death was determined to be internal bleeding caused by internal organ injury with sternum and multiple rib fractures.

Case 2: A 6-year-old male was attacked by a dog, causing diffuse wounds over the entire body. Some of the bites penetrated the abdomen and caused omental bleeding. A large vessel was also ruptured in the right thigh. The cause of death was reported as internal and external bleeding caused by large vessel and internal organ injury, as well as shock from pain caused by the dog bite.

Case 3: A 65-year-old male was found dead near a street after a cattle attack. The external examination revealed widespread abrasion and ecchymosis on the body. The internal examination revealed sternum and rib fractures plus mesenteric bleeding in the abdomen. The cause of death was established as internal bleeding due to visceral organ injury and respiratory insufficiency due to pneumothorax caused by blunt chest and abdominal trauma with sternum and multiple rib fractures.

Case 4: An 83-year-old female was kicked by cattle and died in the hospital the same day as the injury. The external examination revealed diffuse superficial scratches on the body. The internal examination revealed fractures of the skull and symphysis pubis, subarachnoid hemorrhage, and cerebral contusions. The cause of death was reported as cerebral hemorrhage and cerebral destruction caused by general body trauma with skull and symphysis pubis fractures.

Case 5: A 76-year-old male died after being run over by cattle. Rib fractures and seventh cervical vertebral corpus fracture was observed in the autopsy. The cause of death was complications caused by spine and multiple rib fractures.

Case 6: A 74-year-old male with diffuse lesions over the entire body due to dog bites sustained in a dog attack. The internal examination revealed bone fractures and internal organ and large vessel injuries. The cause of death was reported as internal and external bleeding with bone fractures caused by large vessel and internal organ injury because of the dog bites.

A limited number of epidemiological studies about deaths due to animal-related injuries exist in Turkey. This study hopes to motivate the scientific community to look into human casualties as a result of animal attacks in Turkey.

Animal Attack, Blunt and Penetrating Injury, Autopsy

G28 Subdural Hemorrhage as Postmortem Artifact: Characterization of Non-Traumatic Subdural Hemorrhages Identified at Autopsy

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After attending this presentation, attendees will be knowledgeable about a series of cases of presumed artifactual postmortem subdural hemorrhage. Attendees will also recognize artifactual postmortem subdural hemorrhage and understand the factors associated with the development of such collections.

This presentation will impact the forensic science

community by characterizing postmortem subdural hemorrhages, thereby allowing their distinction from antemortem subdural hemorrhages.

Because subdural hemorrhage is typically associated with traumatic head injury, recognition of artifactual postmortem subdural hemorrhage and the characteristics that distinguish it from true antemortem injury is critical for the accurate classification of cause and manner of death. In this presentation, cases of presumed artifactual postmortem subdural hemorrhage are presented and compared to a group of controls.

Cases were defined by the following characteristics: (1) no gross or microscopic evidence of organization; (2) no history of trauma; (3) no evidence of impact sites to the head; (4) no pre-existing organized subdural collections; and, (5) no parenchymal brain hemorrhages or other intracranial lesions. To characterize these hemorrhages, the following information was collected: volume and location of hemorrhage; basic decedent demographic information; cause and manner of death; body position when found; location and degree of lividity at autopsy; underlying medical conditions; brain weight; postmortem interval; and body temperature. To further assess potential factors associated with the development of artifactual postmortem subdural hemorrhage, cases were compared to a control group of 50 decedents without subdural hemorrhage. Controls were randomly selected from decedents with a non-traumatic cause of death performed at the Harris County Institute of Forensic Sciences between July 1, 2012, and June 30, 2013; individuals with intracranial pathology were excluded from the control group.

Ten cases of presumed postmortem artifactual subdural hemorrhage were identified; none were space-occupying and all consisted of thin films over one or both of the cerebral convexities. The cases had the following characteristics: age range of 1 month to 64 years (median: 46.5 years); male gender in 7/10; history of chronic alcoholism in 6/10; hypertensive and/or atherosclerotic cardiovascular disease in 6/10; median brain weight of 1,465 grams; average body temperature at scene of 82.6°F; and average time from last-known-alive to autopsy of 51.7 hours. The control group had the following characteristics: age range of 11 months to 90 years; male gender in 36/50; history of chronic alcoholism in 11/50; hypertensive and/or atherosclerotic cardiovascular disease in 39/50; median brain weight of 1,350 grams; supine body position in 5/10; average body temperature at scene of 87.9°F; and, average time from last-known-alive to autopsy of 28.6 hours. Statistically significant differences observed between cases and controls were the average time from last-known-alive to autopsy (51.7 hours versus 28.6 hours) and history of chronic alcoholism (60% versus 22%).

Artifactual postmortem subdural hemorrhage is a previously undescribed entity. The results of this study suggest that small, non-space-occupying subdural hemorrhages can occur after death. Given the observed association with increased postmortem interval, it is likely that these hemorrhages occur as a result of postmortem breakdown of vasculature in the subdural space, akin to Tardieu spot formation in the skin.

Subdural Hemorrhage, Postmortem Artifact, Case-Control Study

G29 The Difficult Task of Interpreting Cut Marks, Gunshot Wounds, and Ligature Marks on Skin: A Cautionary Note

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PhD, LABANOF, Sezione di Medicina Legale, Dipartimento di Scienze Biomediche, per la Salute, V. Mangiagalli, 37, Milan 20133, ITALY; Marco Grandi, MD, V. Mangiagalli, 37, Milan, ITALY; and Cristina Cattaneo, PhD, Universta Degli Studi Di Milano, Milan, ITALY

After attending this presentation, attendees will gain knowledge concerning the pitfalls of morphological criteria for the evaluation of traumatic skin lesions, for example, in distinguishing injuries due to single- or double-edged blades.

This presentation will impact the forensic science community by revealing the pitfalls in evaluating traumatic skin wounds (cutmarks, stab wounds, gunshot wounds, and ligature marks) on skin.

Gunshot, sharp force, and constriction injuries are common in forensic pathology. Regardless of the type of lesion, the morphological assessment usually performed during autopsy is crucial. Standard "rules" exist for distinguishing between the different types of injuries, such as between an entrance and exit gunshot wound or between a lesion due to a single-edged blade and a double-edged blade. But are these parameters really applicable? If so, how reliable are they and how much interobserver variability among professionals exists?

This study proposes to quantify the diagnostic difficulties in the assessment of sharp force, gunshot, and constriction wounds by macroscopic observation of the lesions. Three questionnaires (one for sharp force, one for gunshot, and one for ligature marks) were given to eight experts (forensic pathologists) and eight non-experts (trainees in forensic pathology) in which the analysis of photographs of different injuries was requested. In the test concerning sharp force wounds, which included 15 photos of single-edged blade and double-edged blade stab wounds as well as cut marks performed with smooth and serrated blades, the observers were requested to state if each injury was performed by a single- or double-edged weapon or by a smooth or serrated blade. For gunshot wounds, the observers were given 15 images and were asked to say whether they were looking at an entrance or exit wound. For constriction marks photographed from soft tissues tied with different types of ligatures, the operators were asked to state if the mark represented in ten photos could be a ligature mark, and if its morphological profile showed signs of concordance with one of eight types of ligatures whose images were provided within the text. In all tests, photographs were high-quality close-ups.

Results showed that for sharp force wounds, the percentage of correct answers was 48% in the group of forensic pathologists, 42% in the group of trainees: in total, 53% of the subjects gave the correct answer on the type of blade, whereas 55% correctly diagnosed the characteristics of the blade. Lesions caused by scissors yielded an even lower success rate, with only 21.6% being correct answers.

In the test on gunshot wounds (whether entrance or exit), the type of lesion was correctly assessed in only 41% of cases, without relevant differences between experts and non-experts. The assessment of gunshot entrance was correct in 49% of the cases, versus 28.7% for exit wounds. For constriction lesions, only 24% of answers were correct. The lesion was recognized in 69% of cases, and the percentage of correct answers was even higher in the trainees' group (74% versus 60% by forensic pathologists). The identification of the type of ligature for specific cases frequently failed, with positive results amounting to 19%.

These results show a simple but important fact: even among expert forensic pathologists open wounds on well-preserved skin can frequently be misinterpreted if classification is only based on external morphology. Theory may give indications of how different stab wounds from single-edged blades and double-edged blades may be, for example, but real-life interpretation may be extremely dangerous and misleading. Basic morphological

assessment should always be performed with caution and backed up by complementary analyses when possible.

Sharp Force Injuries, Gunshot Wounds, Skin

G30 Dissecting Thoracic Aortic Hematoma Masquerading as Blunt Force Injury of the Neck

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After attending this presentation, attendees will be able to discuss the pertinent autopsy findings in cases of cervical compression, describe the pathogenesis of aortic dissection, and elucidate the autopsy findings of aortic dissection which may mimic blunt force injury of the neck.

This presentation will impact the forensic science community by: (1) examining a case in which a natural disease entity masquerades as blunt force injury externally; and, (2) highlighting the importance of having an appropriate index of suspicion when triaging jurisdictional cases for external examination or complete autopsy in order to accurately determine cause and manner of death.

Discoloration of the skin of the anterior and lateral neck may raise suspicion for blunt force injury, particularly cervical compression, especially in the presence of other identifiable injuries in an unwitnessed death. This case presentation will highlight external examination findings in such a situation and correlate them with internal examination findings from autopsy.

A 91-year-old White woman was discovered deceased inside her secure apartment with no significant past medical history and no known next-of-kin. She was last seen alive the day prior by her landlord, who entered with his key the next day and called for emergency services upon finding her on the kitchen floor in a cluttered and unclean apartment. She was clothed in a nightgown and was wearing a necklace and several bracelets. Rigor mortis was weak, and she demonstrated blanching lividity and body warmth was absent. No prescription medications, illicit drugs, or alcohol were found at the scene. With her advanced age, and with the scene investigation suggesting no foul play or trauma, the reviewing pathologist ordered an external examination of the body.

External examination revealed a thin, elderly woman. A band of blue/green skin discoloration spanning from the left neck across the anterior neck and ending beneath the right jaw was identified at initial exam, findings indicating possible cervical compression. Additionally, there were scattered contusions and abrasions on the face, scalp, and extremities. It was decided that an autopsy would be best to determine the etiology(ies) of the cutaneous findings.

Cases of cervical compression will often exhibit intramuscular contusions of the anterior neck muscles. Accompanying features may include cutaneous abrasions and/or contusions of the neck, conjunctival petechiae, and fractures of the hyoid bone or thyroid/cricoid cartilage. With the exception of the blue/green skin discoloration of the neck, none of these were identified in the present case.

Internal examination revealed cardiomegaly with marked atherosclerotic cardiovascular disease and spontaneous rupture of a dissecting thoracic aortic hematoma with tracking of blood along fascial planes of the anterior neck. With the advanced age of the decedent and the thin nature of elderly skin, the blue/green skin discoloration of the neck noted on external examination was attributed to the underlying diffuse extravascular extravasation of blood. Additionally, the abdominal aorta demonstrated an intact

infrarenal aortic aneurysm with marked stenosis due to a 4cm mural thrombus. Internal examination also identified a recent fracture of left anterior-lateral rib 4 and ruled out any lethal injuries. Toxicology testing was negative.

Spontaneous rupture of a dissecting thoracic aortic hematoma occurs most commonly in the setting of atherosclerosis and hypertension. When a dissecting extravascular hematoma of the thoracic aorta ruptures, blood generally accumulates in the mediastinum or chest cavities. The present case illustrates atypical extension of blood into the soft tissues of the neck, mimicking contusion of the anterior neck on external examination.

This case illustrates a natural disease entity masquerading as blunt force injury externally and emphasizes the importance of having an appropriate index of suspicion when triaging jurisdictional cases for external examination or complete autopsy in order to accurately determine cause and manner of death.

Aortic Dissection, Cervical Compression, Autopsy

G31 New Approach to the Traditional English Classification of Trauma and Bone Implications

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After attending this presentation, attendees will obtain knowledge on the current use of various types of trauma injury classifications and terminology, in both Forensic Pathology (FP) and Anthropology (FA). The differences existing in trauma classification among professionals and countries are also highlighted.

This presentation will impact the forensic science community by contributing to the knowledge of various types of trauma classification, apart from basic ones such as blunt, sharp, and projectiles, so that each forensic professional can comprehend the types of documented soft and skeletal injuries. Combined types of trauma like sharp-blunt, penetrating-blunt, and sharp-penetrating will be emphasized.

In English, broad trauma categories, namely blunt, sharp, and projectile, are used to classify, describe, and teach characteristics of injuries; however, the categories are not mutually exclusive of each other. In Portuguese or Spanish *contundente* (blunt), *cortante* (sharp), and *perforante* (perforating/penetrating), previously inspired in early 20th-century French authors and Latin nomenclature, do not express exactly the same meaning as that of English terms. *Contundir* means to beat with force, crushing the tissues with a stick or a rod. For cutting (*cortar*), a knife (used either to cut or penetrate/stab a body), a fragment of glass, a single sheet of paper, or whatever sharp-edged instrument that can actively divide the myofibers is used.^{1,2} The injuries will be wider rather than deep with rectilinear and clean edges. Perforating/penetrating trauma (*perforante*) results from a needle or a conic instrument which has a point and where the depth is superior to the length of the injury. Thus, projectile trauma is a penetrating-blunt injury for many European pathologists, because the projectile (a bullet or a pellet) will injure the skin and soft tissues underneath with a rim of contusion before the penetration. The same pathophysiology applies for injuries that occur from metal spikes, farm forks, and wooden stakes — when the spike is driven through the skin, an inversion will occur with contusion of the margins. In this case,

the designation blunt-penetrating (*perforo-contundente*) is not absolutely new and can be found in European/English literature.³

The point is that the basic tools/weapons (blunt, sharp, and perforating/penetrating) can be used in different and combined forms to produce sharp-blunt (*corto-contundente*), sharp-penetrating (*corto-perfurante*), or penetrating-blunt (*perforo-contundente*) injuries that can be differentiated according to their characteristics, either in soft tissues or bone, that resulted from the speed of the external loading conditions, the point of impact, the resistance of the tissues underneath, etc. The exact meaning of these concepts and their connection with the English terms will be expounded upon along with examples and case studies.

The terminology variants are not important in and of themselves; however, they should be used properly after a good morphologic description of injuries, in an attempt to avoid confusion and misinterpretation of lesions and the suspected weapon, both in autopsy or anthropologic reports, in criminal investigations or in personal testimonies in court.

The final purpose of this presentation is to initiate discussion on this topic and contribute to the clarification of the trauma terminology according to different disciplines and parts of the body that are being studied (bone, skin, etc), always attempting to arrive at the weapon/tool that produced the lesions, although this is sometimes difficult or impossible.

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Classification of Trauma, Sharp-Blunt, Penetrating-Blunt

G32 Blunt Injury vs. Natural Death: A Case of Coronary Arterial Thromboembolism Originating From Mechanical Aortic Valve

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After attending this presentation, attendees will: (1) recognize and assess severity of blunt force head injury; (2) realize the importance of careful review of known clinical details; (3) anticipate and recognize complications of mechanical aortic prosthetic valve; and, (4) be able to identify thromboemboli in major coronary arteries.

This presentation will impact the forensic science community by explaining that blunt injury is one of the most common causes of death in forensic routine work. However, some decedents have coexistence of blunt injuries and severe natural disease. Carefully checking the medical history and performing full autopsy can avoid mistakenly overestimating blunt injury in the cause of death.

The decedent was a 26-year-old G1P1 nursing student at 26 weeks gestation with a high-risk pregnancy. Her medical history is significant for congenital Ventricular Septal Defect (VSD), bicuspid aortic valve, and mild coarctation of the aorta. She underwent VSD repair when she was 8-months-old in 1988. In 1996, she was diagnosed with Discrete Subaortic Stenosis (DSAS) and had surgical removal of a subaortic membrane. As the function

of her aortic valve deteriorated, she had a homograft aortic valve replacement in 1999, which was followed by mechanical aortic valve replacement (St. Jude/Gortex®) in 2006. She was changed from warfarin thromboprophylaxis to subcutaneous low molecular weight heparin for her mechanical aortic valve due to pregnancy.

On the morning of July 5, 2013, she awoke up with numbness in her left arm. She then experienced dizziness, tightness of the chest, and tingling in her arms, followed by stool incontinence once out of bed. At 9:20 a.m., she fell down 15 stair steps on her way to the bathroom. 911 was called and the Emergency Medical Services (EMS) team arrived. She was found conscious, alert, and oriented x3. Upon transfer to the gurney, she suddenly lost consciousness and presented with slow breathing and tachycardia (160bpm). Her transportation was unexpectedly slow due to construction and she was unresponsive upon arrival at the Emergency Room (ER). An Emergent C-section was performed at 10:40 a.m.; however, the mother was pronounced dead at 10:55 a.m. The baby was alive at birth but died one day later secondary to birth asphyxia.

The autopsy revealed minimal blunt force injuries. The skin above and lateral to the right palpebral fissure showed a slightly raised purple/blue abraded contusion measuring 1¼ x 1 inch with a ¼ inch slightly patterned abrasion within the contusion. Petechiae of both the upper eyelids as well as an oval ¾ inch horizontal purple/red contusion of the right upper eyelid were noted. However, no evidence of epidural or subdural hematoma was noted. Sections through the cerebral hemispheres, brainstem, and cerebellum revealed small, rare, periventricular, rubbery, clear-tan plaques measuring 0.6cm. The uncinat gyri and cerebellar tonsils did not demonstrate pressure phenomena. In summary, there was minor trauma of the right orbit, but it was insignificant to cause death.

The dissection of the heart showed a bileaflet St. Jude-type prosthetic aortic valve. A flap-like tan, granular, flat, soft white thrombus measuring 1.5 x 0.6cm was on the inferior surface of the anterior medial leaflet and partially obstructing it. The left anterior descending coronary artery and right coronary artery were patent. However, the mid portion of the circumflex coronary artery was distended and completely blocked by embolic white thrombus. Previous mild post ductal coarctation and surgical closed and healed perimembranous VSD were also demonstrated.

Therefore, the cause of death was determined to be sudden thromboembolic occlusion of mid left circumflex coronary artery secondary to thrombus formation on a mechanical aortic valve prosthesis. The traumatic injuries were incidental, complicating transient cardiac syncope.

Coronary Thromboembolism, Blunt Injury, Prosthetic Valve

G33 Positional Asphyxia and Work-Related Fatality: A Case Report and a Review of the Literature

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After attending this presentation, attendees will understand the importance of the reconstruction of an unusual, fatal occupational accident by a detailed workplace investigation, combined with the evaluation of circumstantial data, and most of all, autopsy findings. The European Statistics of Accidents at Work project defines "fatal accidents at work" as a work-related death of

the victim within a year after the date of the accident. However, the majority of these deaths occur either immediately at the time of the accident or within a few days or a few weeks after the accident.

This presentation will impact the forensic science community by emphasizing how scene investigation, forensic autopsy, and toxicological analyses play a key role in the reconstruction of the dynamics involved in the occupational event, allowing the identification of any legal responsibilities of the worker or the employers.

A 42-year-old man began working on his farmland in the morning. Later in the afternoon, he was found unresponsive with the upper part of his body (head and upper limbs) stuck in the upper and largest aperture (40cm diameter) of an atomizer of a high-volume sprayer tank, with his legs hanging down. The other three smaller apertures (15 cm, 15 cm, and 25cm diameter, respectively) were covered by airtight screw caps. After the body was removed from the tank aperture, resuscitation attempts were unsuccessful and death was pronounced at the scene. The inspection of the tank's interior revealed a few traces of blood and a small quantity of whitish liquid that was submitted to the toxicology laboratory. The external examination of the body revealed discontinuous bruises and abrasions on the skin and soft tissues around the shoulders, some of which encircled the body between the abdominal and thoracic regions. They were characterized by morphological features of the tank aperture in which the victim's body was trapped. The hypostasis/lividity was cyanotic and dark colored. Several petechial hemorrhages were on the anterior neck, facial skin, in the eyes, and on the superior part of the chest. During the autopsy, the gross and histological examinations of viscera showed serosal petechiae, marked venous congestion of the organs, and pulmonary edema. Cardiovascular and respiratory pathologies as well as other pathological diseases were excluded, further confirmed by a review of his medical records. The toxicological analyses of body fluids were negative for alcohol, drugs, and hazardous substances. Chemicals or hazardous substances were not found in the samples of the tank's liquid contents.

The cause of the death was related to positional asphyxia by immobilization of the body and by mechanical impediment which prevented adequate respiratory movements for an extended time. Moreover, due to the victim's position, the upper part of his respiratory tract was trapped in the confined airtight tank space, resulting in deprivation of oxygen. The final determination of the cause of death was essentially based on three criteria: body position obstructing a normal, breathable air exchange; the inability to move to another position; and, the exclusion of other causes of natural or violent death. The information provided by inspectors and police officers who performed the workplace investigation revealed that the victim was using the atomizer for spraying water on vegetables being cultivated in his field. The farmer had climbed on a small wooden stepstool (found close to the atomizer) to reach the upper aperture of the tank for cleaning procedures. The inspection of the machine did not indicate any malfunctions. Considering all the evidence, the manner of death was certified to be an accident at work. The uniqueness of this reported case is confirmed by reviewing the literature.

Occupational Fatality, Positional Asphyxia, Workplace Investigation

G34 Electric or Traumatic Injury? The Role of Histopathological Investigation

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After attending this presentation, attendees will be able to describe the impact of forensic science in cases of electrical injuries.

This presentation will impact the forensic science community by demonstrating the role of histopathological investigation in order to discern electrical and traumatic injuries.

Electrical injury is a relatively rare but potentially devastating multisystem injury with high morbidity and mortality. Examination of the wounds is of primary importance in forensic medicine. Forensic pathologists are often called upon to express an opinion on how an injury was caused and in many cases, microscopic analysis could be beneficial.¹⁻¹¹

The goal of this study is to define the macroscopic and microscopic features of the skin lesions resulting from electrical, mechanical, and thermal trauma in order to formulate a correct differential diagnosis in cases in which these events occur simultaneously. This presentation reports two cases of workers found dead during the course of their work. An autopsy was performed in both cases and the external examinations of the bodies showed skin injuries of uncertain etiological interpretation, further clarified as electrical injuries through histopathological investigations.

Histopathological examination is, therefore, the most accurate survey to detect electrical injury, although it can't provide answers to the immediacy of the event. In these cases, the histopathological examination of skin samples taken during the autopsy allows the examiner to: (1) confirm the macroscopic data emerging from the external examination; (2) demonstrate the compatibility of the lesions with the electrical etiology; and, (3) show the passage of electrical current from the entry point to the exit point.

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Forensic Science, Electrocution, Traumatic Injury

G35 Timing of Blunt Force Injuries in Long Bones: The Effects of the Environment, PMI Length, and Human Surrogate Model

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After attending this presentation, attendees will understand how specific environmental conditions, the length of the Postmortem Interval (PMI), different bone types, and different animal models can influence bone fracture morphology and, consequently, the interpretation of blunt skeletal trauma as having occurred perimortem or postmortemly.

This presentation will impact the forensic science community by increasing understanding of the timing of blunt force injuries in long bones with new data concerning the analysis of fractures in long bones under different postmortem conditions, using the Fracture Freshness Index (FFI). Moreover, new data concerning variations in fracture morphology between different animal models and bone types is presented, emphasizing the complexity of extrapolations to the human context.

The timing of blunt force trauma in human skeletal material is a critical issue in forensic pathology and anthropology. However, there is still limited knowledge as to how fracture morphology is influenced by specific environmental conditions, by the length of the postmortem interval (PMI), by different bone types and by different animal models. The goal of this study is to evaluate the influence of the type and duration of the postmortem environment in interpreting blunt skeletal trauma as perimortem or postmortem, based on comparisons of fracture morphology from long bones with different postmortem intervals and decomposition environments while simultaneously assessing variations in fracture characteristics between different bone types and species. Fresh limb segments from pigs and goats were used in this study and were sequentially left to decompose under three different environmental circumstances (ground surface, buried, and submerged) during a total period of 196 days, after which all sets of limb segments (each with different PMI) were fractured together with a fresh set. Fractured bones (total n=325; pig tibia=110; pig fibula=110; goat metatarsals=105) were assessed macroscopically and classified according to the FFI. Climatic data for the location of the experiment was collected. Statistical analysis included descriptive statistics, correlation analysis between FFI and PMI, Man-Whitney U tests for comparisons of FFI medians for different PMIs and linear regression analysis using PMI, mean pluviosity, and mean temperature as predictors for FFI. Surface samples presented increasing FFI values for each PMI increment, with positive correlation for all studied bone types, the same observed in submerged samples, except for pig tibia. Median FFI values for surface samples with PMI=0 could be statistically differentiated from PMI=56 days or above. Buried samples presented no significant correlation between FFI and PMI as well as no statistically significant linear regression

models. Linear regression analysis of surface and submerged samples suggested differences in FFI variation with PMI between bone types, although it failed to show statistical significance. When adding climatic data to the surface regression models, PMI was no longer a predictor of FFI. When comparing different animal models, linear regressions seemed to suggest greater increases in FFI with increasing postmortem period in pig samples compared to goat samples in both surface and submerged environments, but they failed to reach statistical significance. No differences were found between environments except for buried versus submerged metatarsal goat samples and surface versus buried or submerged tibia pig samples. FFI seems to have a weak association with PMI, possibly due to the slow-rate fracture morphology changes with increasing PMI, and it seems to be affected by various factors, such as different bone types, decomposition environment, and climatic factors. Nonetheless, it does show some discriminating power in fracture morphology during the early postmortem period. The apparent variation between bone type cautions against current experimental studies, as extrapolations to the human species can be challenged. The present study demonstrates the potential of the FFI in reflecting fracture morphology changes over time; however, perimortem or postmortem fracture diagnosis based on the FFI seems to be extremely difficult.

Bone Fracture Morphology, Postmortem Interval, Postmortem Environment

G36 Laryngeal and Hyoid Bone Trauma: Unusual Injuries in a Suicide Jump Into the Water

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After attending this presentation, attendees will understand how important a complete autopsy is, including paying special attention to neck structures, the histological and toxicological exams, the death scene investigation, and analysis of the circumstances in cases of laryngo-hyoid trauma as a result of presumed suicide by jumping from a height into the water, especially to exclude a possible homicide.

This presentation will impact the forensic science community by showing how useful a formaldehyde fixation of the neck organs can be, prior to their dissection, allowing an easier dissection of this area and better identification of the injuries and how important the integration of all information to determine the manner of death.

In forensic contexts, falls from great heights are suicidal in most cases. However, accidents can occur as well as homicides. The presence of neck injuries, particularly laryngeal and hyoid bone trauma, are not common, especially when water impacts are involved. These findings always raise the possibility of a previous homicidal strangulation.

In the forensic literature, reports of laryngo-hyoid trauma after falls from a height are not frequent. The majority of the cases concerned impacts of the victims on the ground and little information is available for impacts on water surfaces. Even for impacts on the ground, some studies found this trauma in no more

than 5% of the cases. In reviewing 65 cases of suicide by jumping from the Bosphorus Bridge in Istanbul, in only two cases were laryngochoyoid fractures found. Another study of 169 cases involving suicide jumps from the Golden Gate Bridge in San Francisco didn't report any case of laryngochoyoid trauma.

Even if in suicidal situations laryngochoyoid fractures can be present, their interpretation should be taken with considerable caution, bearing in mind the possibility of a previous homicide attempt.

This study presents a case of a 56-year-old businessman with financial problems. His car was found parked on a bridge with the emergency flashers engaged. A witness saw a body floating in the river about 20 meters downstream from the bridge. The person was not signaling for help, and the body sunk after a few minutes. The height of the bridge to the river bed was about 40 to 50 meters. The body was found 24 hours later.

During the autopsy, the external examination found a large ecchymosis in the left ear, little ecchymosis in the left orbital region, nose, anterior view of the left arm and left leg, and transversal and parallel abrasions on the thigh.

The internal examination showed more significant injuries, namely blood infiltration of the frontal and temporal regions on the left side, of the muscles of the face on the same side, on the right side of the neck, and also of the proximal pre-cervical muscles. The large horns of the hyoid bone as well as the superior horns of the thyroid cartilage were fractured and also surrounded by a significant hemorrhage. Vertebral cervical fractures and fractures of the ribs and sternum were present. Small tears in the intimae of the descending aorta, with blood infiltration, and contusion of the heart and pancreas complete the traumatic findings. Cerebral and pulmonary edema as well as aqueous foam in the bronchia were also found.

The neck organs were removed *en bloc* and subjected to formaldehyde fixation. A careful dissection allowed a better identification of the fractures, edema of the aryepiglottic folds, and haematomas at the pharyngeal and esophageal mucosa. Histological exams were performed as well as toxicological, which were negative for alcohol, drugs, and pesticides. The first exams confirmed the vitality of the injuries and the presence of alveolar lesions, type "*emphysema aquosum*," in the lungs and diffuse vascular congestion, consistent with drowning.

After the autopsy, a farewell note was found inside the car of the victim. As the police didn't find anything suspicious during the investigation, the case was classified as a suicide.

This study underlines how, in situations of laryngochoyoid fractures resulting from impact on water surfaces, a careful autopsy combined with histological and toxicological exams and the information from the death scene investigation are crucial in determining the manner of death and excluding homicide.

Fall, Laryngochoyoid Fractures, Suicide

G37 Gorham-Stout Syndrome: A Presentation of Two Cases and a Review of the Literature

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The goal of this presentation is to investigate cause of death in Gorham Stout Syndrome (GSS). Prognosis is most dependent

on the site of involvement, extent of disease, and complicating factors. It remains a fact that several cases involve the thoracic duct and the death can occur as a sequelae of malnutrition and lymphopenia. Chylothorax is a serious complication and there are no uniform therapeutic guidelines to follow. The very low incidence of this syndrome as well as the chylothorax clinical presentation and the poor understanding about the etiology make diagnosis difficult.

This presentation will impact the forensic science community by describing morphological features of a rare syndrome and by confirming the importance of detailed postmortem forensic investigations as a substitutable way to improve diagnostic criteria and therapy.

GSS is a rare disorder characterized by progressive osteolysis that leads to the disappearance of bone. Lymphovascular proliferation causes the local destruction of bony tissue. Children and teenagers, without differences between males and females, are usually affected. The first clinical symptoms are pain, tumefaction, and spontaneous fractures of bones. The progression of osteolysis may stop at any moment, but it often evolves with a complete waste of the bone, with the replacement of a fibrous band. Less than 200 cases are documented in the literature. The outcome is poor in patients with soft tissue involvement, with lesions of the axial skeleton and chylothorax. Twenty-five (17%) of patients have been reported to have chylothorax as a complication, according to the last review of the literature.

This presentation reports two fatal cases of GSS.

Case 1: A 9-year-old child affected with GSS was referred to the local Emergency Department (ED) because of severe dyspnoea. Respiratory Function (RF) was 36/min with a rough breath sound. Oxygen saturation was 88%. Tachycardia was recorded with Heart Failure (HF) at 140/min. Blood Pressure (BP) was unremarkable. Respiratory acidosis was detected at lab tests (pH 7.28, pO₂ 30.3, pCO₂ 57.9, HCO₃ 22.0). Complete opacification of the whole left lung with right pleural effusion were observed on an X-ray of the chest. Therapy with an oxygen mask was immediately started and the child was moved to the intensive care unit because of acute respiratory failure. He was unconscious, cyanotic, tachypneic with oxygen saturation at 78%, and 500cc of fluid was drained from the left thorax. Dopamine infusion was quickly carried out; however, after an episode of severe bradycardia, the child collapsed. A hospital autopsy was performed the day after the child's death. The child was 139cm in length and weighed 27kg. At external examination, asymmetry of the head due to osteolysis in the parietal, temporal, and mandibular regions was described. Chylothorax and massive pulmonary edema was described at autopsy. Gross examination of the heart was unremarkable. The histological findings of the lungs were suggestive of diffuse pleural effusion characterized by large, thin-walled, and variably sized lymphatic vessels. The immunohistochemical staining method revealed vascular proliferation in the parietal and visceral pleura. Acute respiratory failure in GSS was indicated as the cause of death following a mediastinic syndrome with metabolic and respiratory acidosis and inspiratory dyspnoea.

Case 2: A 5-year-old child with a history of mediastinic syndrome affected by GSS was admitted to the local pediatric surgery department due to high fever and left chest pain. Chylothorax was observed in the chest and 600cc of lymphatic fluid was seen in the left thorax. Fatal bradycardia occurred suddenly. A hospital autopsy was performed and the external examination was unremarkable. Massive cerebral edema was recorded with white foam on the main bronchi. Yellowish liquid in the left thorax was detected. Disarray of the myofibers and multiple foci of contraction band necrosis were observed at the microscopic study of the heart. Interstitial myocardial edema and lymphangectasia were also described. Pleural lymphatic vessel proliferation with focal pulmonary edema was recorded as well as broncho-pneumonia

and bronchiolitis. The histological examination of the liver revealed dilatation of lymphatic vessels. The immunohistochemical staining method revealed generalized vascular proliferation of all organs. Acute cardiac failure in GSS was indicated as cause of death.

Gorham Stout Syndrome, Osteolysis, Child Disease

G38 Infant Death Investigation: A Retrospective Study From 2002 to 2011

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The goals of this presentation are to determine the incidence of the principal causes of infant deaths, analyze the risk factors and precipitating circumstances, assess the importance of a deep forensic investigation for the determination of the cause of death, and suggest possible prevention measures.

This presentation will impact the forensic science community by describing how the implementation of multidisciplinary approaches and protocols for the classification of all sudden and unexpected infant deaths is absolutely fundamental. Placing infants in a supine position to sleep, avoiding tobacco exposure in pre- and postnatal periods, encouraging breastfeeding, and avoiding bed sharing are some of the prevention measures which may be largely implemented.

A retrospective study was conducted to analyze the number of infant death (under age one) cases which occurred in Northern Portugal during a 10-year period.

The most common presentation of infant death is Sudden Unexpected Death in Infancy (SUDI), defined as deaths in infants under one year of age that occur suddenly and unexpectedly, and whose cause of death is not immediately obvious prior to investigation. According to the Confidential Enquiry into Stillbirths and Deaths in Infancy (CESDI) guidelines, deaths are classified within SUDI if they occur between 7 and 365 completed days of life and fulfill the following criteria: deaths that were unexpected and unexplained at autopsy; deaths during an acute illness that was not recognized as life-threatening; deaths due to an acute illness of less than 24h duration in a previously healthy infant (or death after this period if life had only been prolonged by intensive medical care); deaths from a pre-existing occult condition; and, deaths from any form of accident, trauma, or poisoning.

Thus, SUDI comprises a heterogeneous group, including deaths in which a careful review of the death scene and a meticulous postmortem examination will disclose a cause of death and those which will remain unexplained even after such examination.

After this thorough case investigation, infant deaths may be explained in cases with known and internationally accepted cause of death, but the related diseases were either lacking serious preceding symptoms or were not recognized. These include natural deaths due to acute medical illnesses, such as pulmonary injury related to ingestion of gastric content and infections (pneumonia and viral myocarditis) but also violent deaths (accidental or homicide), such as suffocation and poisoning.

In order to achieve the goals of this study, the forensic autopsy reports of infant deaths that occurred between January 2002 and December 2011 at the North Branch of the National Institute of Legal Medicine and Forensic Sciences (Portugal) and its offices were reviewed.

The analysis included a total of 103 sudden and unexpected deaths in infancy (SUDI), and the cases were divided

in neonatal (17 cases, 16.5%) and postneonatal (86 cases, 83.5%). Infant deaths were divided into explained causes (85 cases, 82.5%), which included disease-associated causes of death (68 cases, 66%), as well as violent causes (17 cases, 16.5%), and unexplained causes (18 cases, 17.5%), classified as Sudden Infant Death Syndrome (SIDS).

Infant Death Investigation, Causes of Death, SUDI

G39 The Choking Game: A Deadly Game — Analysis of Two Cases of Self Strangulation in Young Boys

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After attending this presentation, attendees will know about the choking game, which is more frequent than expected and often goes unrecognized.

This presentation will impact the forensic science community by showing the importance of an accurate analysis of the circumstances of death, an in-depth inspection of places and things, and a careful autoptical and toxicological examination to do a correct diagnosis of a death by choking game.

The goal of this presentation is to provide information to show how cases of the “choking game” are more frequent than expected despite the fact that the phenomenon often goes unrecognized. The “choking game,” also called “jeu du foulard” in French-speaking countries, is defined as a self-strangulation or strangulation by another person with the hands or a noose to achieve a brief euphoric state caused by cerebral hypoxia. This provides a dizzy sensation, which is described as “cool.” Loss of consciousness may occur with potential injury from subsequent falling and/or due to hypoxic injury. Death may also occur, but young people do not seem to understand the danger of this behavior. It can involve both males and females, and the age range is 9-19 years, with an average of 13 years of age. Choking game participation is often associated with some health risk categories, such as poor mental health, substance use, exposure to violence, sexual activity, and gambling. These deaths are often classified as suicides or accidental deaths, without considering the possibility that they are a result of a deliberate self-asphyxiation in order to derive pleasure, which then turns into a deadly game. Death scenes and autopsy findings in suicides and asphyxial games are similar, so it is important to know the problem in order to better investigate. That’s why further studies are necessary in order to be able to recognize and adequately prevent this deadly game which can be difficult to identify. This presentation will impact the forensic science community by providing key information that can help correctly evaluate asphyxia cases in young persons, to understand if it’s suicide, accident, or a choking game, by presenting two cases involving two young boys who died as a result of a self-strangulation.

In April 2009, an adolescent 11-year-old boy was found by his stepfather, hanged from a bunk bed in his bedroom by a scarf made into a noose. His stepfather removed the ligature and began cardiopulmonary resuscitation. The boy was transported to the local hospital where he was pronounced dead one hour later. An accurate inspection was made, including the analysis of the young man’s computer. The results of this inspection, autopsy findings, and the testimonies of parents and teachers made it possible to confirm the death was a result of a “choking game.”

In June 2012, another adolescent boy, 15 years old, was

found with a scarf looped about his neck, suspended on the bunk bed, which was 103cm (3.4 feet) high, by his sister who went to his room to check on him because she hadn't seen him in three hours. The mother began cardiopulmonary resuscitation while his sister called the ambulance, but the boy was pronounced dead by physicians.

These two cases show the importance of a careful knowledge of this dangerous "game," which is widespread among young people all over the world. An accurate analysis of the circumstances of death, an in-depth inspection of places and things, and a careful autoptical and toxicological examination are essential in identifying the circumstances and cause of death and for the diagnosis of death due to strangulation during a "choking game."

Choking Game, Self-Strangulation, Asphyxial Game

G40 Hand, Foot, Mouth, ... And Medulla: Fatal Encephalitis in a Toddler Associated With Enterovirus 71 Infection

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After attending this presentation, attendees will understand the presentation, pathophysiology, and epidemiology of the emerging syndrome of Enterovirus 71-(EV 71) associated encephalitis in children.

This presentation will impact the forensic science community by elucidating the situations in which this condition should be considered and how identification at autopsy may be maximized. The significance of these cases to the public will be discussed.

A 17-month-old, previously healthy, female toddler presented to a Children's Hospital with fever, pallor, vomiting, poor perfusion, and shallow breathing. Initially tachycardic and self-ventilating, she abruptly developed bradycardia and deteriorated into cardiopulmonary arrest, from which she could not be resuscitated. During this brief terminal encounter, clinicians noted abnormal ocular findings, specifically ptosis, midline deviation, and nystagmus.

This child had been evaluated twice during the day preceding her death for suspected respiratory tract infection. No rashes or neurological findings were documented.

Gross autopsy findings included serous effusions, increased lung weight, and cerebral edema. The bladder was distended with urine. The lungs showed intra-alveolar edema and lymphocytic interstitial pneumonitis. Enterovirus was recovered from a lung swab. Examination of the fixed brain by a neuropathologist showed moderate symmetric brain swelling with dusky gray/brown discoloration, and congestion in the brainstem. Microscopy revealed meningoencephalitis, most severe in the caudal brainstem with focal necrosis, less severe in the cerebellate dentate nucleus, and relatively mild in the cerebrum (confined to the motor cortex, globus pallidus (inner segment), basal forebrain, hypothalamus, and inferior thalamus/subthalamus), typical of EV71 infection. No viral inclusions were identified.

The Enterovirus was subsequently sub-classified as EV 71. At least 3 children have died of EV 71-associated encephalitis in New South Wales this year, all identified following autopsy.

There is a clear association between Hand, Foot, and Mouth Disease (HFMD) and EV 71 and Coxsackie A virus 16, and a clear association between EV 71-associated HFMD and encephalitis. There is no strong evidence of a particular associated strain or recombination between strains of EV or Coxsackie viruses.

HFMD is worldwide in distribution and outbreaks with associated encephalitis have been documented, although reports from Asia predominate. HFMD is a very common, highly infectious childhood disease with a highly variable presentation (non-specific, respiratory, or gastrointestinal); classically there is a vesicular rash on the hands and feet, or herpangina (vesicles within the mouth, often the posterior oropharynx) although the exanthema is often absent.

Possibly 6% of infected children will develop encephalitis. The prototypical case will be a child under three years of age with a brief, non-specific viral illness, with or without exanthema, in the summer or fall; deterioration will be rapid, with onset of any of a large variety of neurologic signs, although acute flaccid paralysis (sometimes monoplegia), myoclonic jerks, and cranial neuropathies are frequent. The mechanism of death is usually neurogenic pulmonary edema.

Forensic pathologists need be vigilant, as presentation may be non-specific and could be mistaken for Sudden Infant Death Syndrome (SIDS). Viral studies should be obtained, especially from the upper respiratory tract and rectum; cerebrospinal fluid is less reliable. Herpangina should be specifically sought, since the posterior oropharynx may be a "blind spot" in a routine autopsy. Ideally, the brain should be fixed and obviously examined with care, for it is the site of definitive pathology. If Enterovirus is recovered in the appropriate setting, the laboratory should be asked to sub-classify it. These cases are likely to be scrutinized by child mortality and public health authorities.

Although rare, these cases attract significant public interest and concern. It is unexpected in developed countries for children to die of an infectious disease, and in so rapid a fashion with dramatic neurologic signs; the presentation is ominously reminiscent of polio. The prodrome is indistinguishable from the usual frequent childhood viral illnesses. The public must invoke hygiene measures and isolation. Restriction from day care and support of children who survive with neurological impairment are economic burdens. Clinicians who see cases in the early stages may be considered culpable for a subsequent death. Trials of a promising vaccine are ongoing in China.

Enterovirus, Encephalitis, Children

G41 Fatal Tube Feeding Syndrome in a Young Child

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The goals of this presentation are to: (1) list the types of dehydration based on the relative losses of salt and water and on the composition and volume of intake; (2) list causes of hypertonic dehydration; and, (3) estimate free water deficit based on a child's weight and serum (or vitreous) sodium.

This presentation will impact the forensic science community by increasing awareness of the potential fatal consequences of enteral feedings with elemental hyperosmolar solutions in young children.

Tube feeding syndrome was first described in the 1950s as a cause of hypernatremia and hyperosmolar dehydration in adults, especially those with head injury or who were otherwise unable to communicate. Subsequently, it was recognized that decreasing the osmolality of the solutions introduced via enteral tube feeding, in conjunction with increasing free water via parenteral or enteral methods, was successful in correcting or avoiding most electrolyte

abnormalities. There has also been discussion concerning the importance of hyperosmolar dehydration causing a significant increase in morbidity and mortality in the pediatric population, especially if not corrected.

This study presents the case of a 2-year-old child born at 25 weeks estimated gestational age who developed cerebral palsy, chronic lung disease of prematurity with associated pulmonary hypertension, and retinopathy of prematurity. She exhibited developmental delay and feeding intolerance secondary to oral aversion for which she had received a gastrostomy tube.

She was admitted for feeding intolerance, vomiting after feeding, and failure to thrive secondary to complications of prematurity and for evaluation of gastro-jejunosomy tube placement. Initial evaluation revealed a 7.7kg female with a body length of 78.4cm, placing her below fifth percentile growth for both weight and length corrected for age. Admissions laboratory results included serum sodium 137mmol/L, chloride 103mmol/L, urea nitrogen 8mg/dL, and creatinine 0.33mg/dL. Nutrition and gastroenterology consultations recommended against gastro-jejunosomy placement and feedings were titrated over the first day with her home mixture of hyperosmolar (850mOsm/kg water) essential amino acid solution with high-caloric powder supplement. Multiple attempts at intravenous access were unsuccessful. Additional free water, either parenterally or enterally was never provided, and with oral aversion and developmental delay, the patient was unable to communicate thirst.

She had gone to sleep uneventfully the evening prior to death (five days after admission) and nursing notes at 4:00 a.m. on the date of death (six days after admission) cited no abnormality, with mother and child sleeping peacefully in the room. At 8:30 a.m. on the sixth day after-admission vital check, a nurse noted that the child had a pale ashen color, was cool to the touch, and was stiff without pulse or respiration. She was emergently coded for asystole and eventually pronounced at 9:06 a.m.

At autopsy, the ocular conjunctival membranes along with lips, anterior tongue, and buccal mucosa were dry and pale. Her peritoneal and pleural cavities were dry and tacky. The trachea was dry and the lungs demonstrated atelectatic areas comprising approximately 25% of the right lung's lower lobe, 10% of the right lung's upper lobe, and 50% of the left lung's lower lobe. The gastrostomy tube was in place and well seated with residual enteral feeding solution in the stomach. Minimal urine volume was observed within the bladder. Encephalopathy of prematurity was confirmed on neuropathology review with periventricular leukomalacia, periventricular hemosiderin deposition, cerebellar hypoplasia, and mild hydrocephalus.

Due to the appearance of dehydration at autopsy vitreous electrolyte analysis was completed demonstrating sodium 168mmol/L, chloride 151mmol/L, urea nitrogen 49mg/dL, and creatinine 1.33mg/dL. Urine specific gravity was 1.024. Testing of the enteral feeding solution exhibited osmolality 925mOsm/kg with sodium <100mmol/L and chloride <50mmol/L.

Hyperosmolar dehydration is a well-documented cause of morbidity and mortality in pediatric populations, especially when combined with other conditions. With the rise in survival rate of premature neonates and associated neurologic complications, it is important to recognize the danger of hyperosmolar feeding solutions and how they contribute to feeding tube syndrome in the growing pediatric population that requires them. The medicolegal investigator must be able to recognize the sequelae of dehydration at the time of autopsy and correlate with appropriate antemortem and postmortem electrolyte studies to detect such derangements.

Forensic Science, Tube Feeding Syndrome, Hyperosmolar Dehydration

G42 Concealment and Denial of Pregnancy and Neonaticide: A Report of Four Cases

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The goals of this presentation are to describe and discuss four cases of neonaticide in which the pregnancy was denied or concealed by the mother.

This presentation will impact the forensic science community by demonstrating that concealment and denial of pregnancy are definitively suspicious factors when investigating a neonaticide.

Neonaticide is the deliberate killing or homicide of a child within 24 hours of birth. The victim of neonaticide is often the product of an unwanted pregnancy. The perpetrator is usually the young mother, described as immature and unmarried. In virtually every case, the mother is the lone perpetrator. The birth and the killing are unwitnessed and the killing occurs immediately after the birth.

Seven cases of neonaticide were identified during the period 2009-2012 at the Institute of Forensic Sciences of Puerto Rico. Four of these cases involved the birth of a live infant to a young mother who had reportedly concealed or denied her pregnancy.

Case 1: A 21-year-old first-time mother abandoned her baby in the toilet of her house after the delivery. She told the investigators she did not know she was pregnant. The baby was found dead in the toilet with only part of the torso in contact with the water. Autopsy disclosed a full term 8-pound, 21-inch male neonate with attached placenta. The newborn was alive and healthy at birth. The body had no signs of trauma. Cause of death was ruled as neonatal asphyxia and manner of death as homicide.

Case 2: A 16-year-old woman had hidden her second pregnancy from her close relatives. Later, she confessed that she was in her 30th week of pregnancy. She delivered a baby girl in the bathroom of her house, immediately killed and disposed the body of the newborn in a plastic bag, and cleaned the scene. The autopsy disclosed that the 7-pound, 19.7-inch neonate was born alive and had no macroscopic congenital abnormalities. Trauma of the head included subdural and subarachnoid hemorrhages. The face had congestion around the nose and adjacent to the mouth. There were petechiae over the eyelids and abundant froth at the nostrils. The neck had hemorrhagic infiltrates of the strap muscles. The cause of death was ruled as severe cerebral trauma and asphyxia by suffocation and strangulation.

Case 3: A 21-year-old first-time mother gave birth to a baby boy in the shower area of the family bathroom. She stated that she did not know she was pregnant and assumed she was constipated. The baby was found still alive and was taken to the emergency room and died hours later. Autopsy revealed that the full term, 6-pound, 21.7-inch baby was alive and healthy at birth. Contusions were present all over the scalp and abundant subgaleal hemorrhage was also noted. The cranial vault had multiple complex and diastatic fractures. Subdural and subarachnoid hemorrhages were over the cerebral hemispheres. Recent cerebral contusions and diffuse vascular injury were also present. The left eye had retinal and optic nerve hemorrhages. The cause of death was ruled as severe head trauma and manner of death was homicide.

Case 4: A 21-year-old nursing student and mother of a child told the police that she did not know she was pregnant after she gave birth to a baby girl in the bathroom of her house. The dead body was found in the shower area wrapped with a shower curtain. The autopsy revealed that the 7-pound, 20.4-inch baby girl was alive and healthy at birth and had signs of strangulation and suffocation.

Abundant subgaleal hemorrhage and complex fractures of the cranial vault were also present. Diffuse subarachnoid hemorrhage was over the brain. The cause of death was ruled as severe head trauma and asphyxia by strangulation and suffocation and manner of death was homicide.

Often when presented with a suspected neonaticide, investigators will note whether or not the mother attempted to conceal or deny the pregnancy and/or the delivery of the child. Such concealment is definitely a suspicious factor. The above-presented cases demonstrate in great part the spectrum of investigative and pathological findings in neonaticides.

Neonaticide, Pregnancy, Cause and Manner of Death

G43 Adolescent Suicide: A 30-Year Retrospective Review

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After attending this presentation, attendees will understand the trends relative to adolescent suicide over a 30-year period in the Miami-Dade County District of the Florida Medical Examiner System.

This presentation will impact the forensic science community by making known the problem of adolescent suicide including demographics and trends of suicide modality over time.

Suicide is the tenth leading cause of death across all ages in the United States and has been described as the third leading cause of death in adolescents and young adults 10-24 years of age. Though suicide among adolescents is rare, it is rising in some jurisdictions and becoming a major public health problem. This study reviewed case materials for suicides in children younger than 18 years of age that were investigated by the Miami-Dade County District jurisdiction from 1982 to early 2013. Over these approximately 30 years, there were 187 suicides committed by children younger than 18 years of age compared to 7,661 suicides committed by adults during the same time period. Adolescent suicides comprised 2.4% of suicides overall and ranged from 0.5-6.3% over the years of the study. On average, there were six adolescent suicides per year. The highest number of adolescent suicides was in 1988 when there were 14; the lowest number was in 2006 when there was a single case. The mean and median ages of the decedents were 15.23 years and 16.00 years, respectively. The age range of the decedents was from 8 to 17 years. Of the adolescent suicides, 78% were male and 22% were female. Forty-two were Black and 140 were White. One was of unknown race. Four were described only as Hispanic. Suicide notes were found in 20 cases (10.8%).

Gunshot Wound (GSW) was the most common method of committing suicide, comprising 57% (106). Eighty-five boys and 21 girls committed suicide by gunshot. Asphyxia, including 48 hangings and one suffocation with a plastic bag, was the second most common modality, comprising 27% of suicides. Forty-three boys and six girls died by asphyxia. Other modes included poisonings (13), blunt trauma from falls from height and an intentional collision with a train (9), drowning (9), and thermal injury (1). In the majority of the traumatic suicides, no drugs were detected.

Of the 106 cases of adolescent suicide by GSW, 12 were the result of Russian roulette and three were alleged to have been "playing" with a gun. From 1982 through 2005, GSW consistently outpaced hanging as the most common suicide modality; however, from 2010 through early 2013, there were only four GSW suicides compared to 19 other modality types including 15 hangings.

Girls used poisoning as a suicide modality more frequently

than boys. The 13 deaths by poisoning included ten girls and three boys. These suicides by poisoning were confirmed by toxicological analysis.

This study confirms the reported gender disparity in suicide as well as the disproportionate use of guns in boys and poisoning in girls. The risk factors for suicide in adolescents are a history of a previous attempt, depression or mental illness, alcohol or drug abuse, a stressful life event, and incarceration. The vast majority of these cases had one or more of these risk factors. Interestingly, in this jurisdiction, despite the increased availability of firearms, the number of suicides by GSW has been declining among adolescents while the number of suicides by asphyxial means has been rising. Prevention measures focused solely on restricting access to the most lethal means, i.e., firearms, are likely to have limited success since the choice for the method of suicide appears to be most influenced by what is readily available and convenient.

Suicide, Adolescent, Pediatric

G44 Congenital Mesenteric Fenestration With Hernia Causing Intestinal Obstruction and Sudden Death of a Child

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After attending this presentation, attendees will understand the importance of full forensic evaluation in cases of children with sudden death following gastrointestinal complaints.

This presentation will impact the forensic science community by illustrating a rare but important presentation of internal small bowel herniation which led to sudden death in a child. The presentation alerts the forensic community about a rare cause of sudden death in the pediatric population, its preceding symptoms and presentation, and reviews the available literature.

An internal hernia is a protrusion of bowel through an orifice in the peritoneum or mesentery.¹ Transmesenteric hernias are internal hernias caused by a congenital defect in the mesentery.² As no hernia sac is involved, a considerable length of bowel can protrude through the defect.³ Despite the congenital nature of the mesenteric defect, this phenomenon can present at any age, with adults making up most of the cases reported.⁴

This presentation reports a rare case of a four-year-old girl who presented with abdominal pain and vomiting and became unconscious a few hours later. The girl was taken to the hospital and died despite attempted resuscitation. She had no previous medical or surgical history. A computed tomography of the abdomen showed multiple dilated loops of small bowel consistent with either obstruction or ileus. The laboratory findings included leukocytosis and metabolic acidosis. A subsequent autopsy revealed that the small bowel had herniated through a small congenital fenestration in the mesentery leading to incarceration, hemorrhagic infarction, and intestinal obstruction. The mesenteric defect measured approximately 1.5 centimeters. The herniated segment of small intestine measured approximately 40 centimeters and was microscopically necrotic. Herniation through a congenital mesenteric defect is an uncommon and extremely serious cause of intestinal obstruction. Most internal hernias occur postoperatively, resulting from incomplete closure of surgically created mesenteric defects.⁵ An acute intestinal obstruction with bowel entrapment in the absence of an external hernia and with no history of previous surgical procedures should suggest the possibility of an internal hernia.⁶

This case illustrates the difficulty of diagnosing incarcerated

internal hernias due to nonspecific accompanying symptoms and its rapid progression to intestinal infarction and death. Immediate surgical intervention is needed to prevent subsequent mortality. It is important that sudden unexpected deaths in children undergo full forensic evaluation to establish the precise cause of death.

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Mesenteric Hernia, Bowel Obstruction, Children

G45 Sudden Death Due to Segmental Arterial Mediolytic Involving the Right Coronary Artery

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After attending this presentation, attendees will learn to recognize segmental arterial mediolysis as a potential cause of sudden cardiac death and to distinguish it from other causes of coronary artery dissection.

This presentation will impact the forensic scientific community by focusing attention on nonatherosclerotic causes of lethal coronary artery disease in general and on one potentially overlooked cause, segmental arterial mediolysis, specifically.

A 65-year-old woman with a past medical history significant for hypercholesterolemia and hypertension presented approximately three months prior to death with a chief complaint of rib pain. Radiographic and laboratory studies and a bone marrow biopsy were consistent with multiple myeloma. She was treated with bortezomib, dexamethasone, zoledronic acid, and lenalidomide and tolerated it well. On the day of her death, she complained of back pain, and late that night she suddenly became pale and unresponsive. Emergency medical personnel found her in ventricular fibrillation, initiated resuscitative efforts, and transported her to the emergency department. Physical examination revealed a pulseless, unresponsive, afebrile, slightly overweight (BMI of 26) woman with an unremarkable body habitus. Laboratory results were significant for metabolic acidosis and a troponin I value that was elevated but below the range considered consistent with acute myocardial infarction. The clinical cause of death was given as hypoxia due to metabolic acidosis due to multiple myeloma.

Autopsy revealed cardiomegaly and left ventricular hypertrophy. The left-sided coronary arteries showed minimal to mild coronary artery disease. The posterior interventricular branch of the right coronary artery appeared thrombosed, and

the gross impression was of probable coronary artery disease with superimposed thrombosis. Histology revealed instead a right coronary artery dissection with medial hemorrhage and luminal occlusion. The surrounding myocardium showed histologic evidence of recent infarction. Changes similar to those seen in the right coronary artery were incidentally found in a medium-sized branch of the splenic artery. Neither of the affected vessels showed atherosclerotic changes, vasculitis, or giant cells. Both showed disruption of the elastic lamina highlighted by elastic stains. An incidental small pulmonary embolus was found in the right middle lobe. The aorta and its great branches showed trace atherosclerotic changes. No histologic evidence of myeloma was found in the marrow.

As a group, nonatherosclerotic coronary artery lesions are a rare cause of sudden death. The list includes congenital coronary artery anomalies (e.g., hypoplasia and anomalous origin), coronary arteritis (e.g., polyarteritis nodosa, giant cell arteritis, Kawasaki disease, and luetic arteritis), coronary artery spasm, and mechanical obstruction resulting from coronary artery dissection. Spontaneous Coronary Artery Dissection (SCAD) is most often seen in women younger than 45, often in the peripartum period, and typically in the left-sided coronary arteries. Dissection has also been associated with arteritis, connective tissue disorders such as Marfan syndrome and Ehlers-Danlos syndrome, cocaine use, and strenuous exercise. The findings in the present case are consistent with Segmental Arterial Mediolytic (SAM), another rare and potentially under-recognized cause of coronary artery dissection and sudden cardiac death.

First described in 1976, SAM is characterized by destruction of the media of small- to medium-sized arteries with loss of the elastic lamina and separation of the media from the adventitia. There is no evidence of significant inflammation or atherosclerotic change. The resulting structural compromise can lead to aneurysm formation, dissection, or rupture of involved vessels with infarction of supplied tissue. While SAM most often involves abdominal arteries and presents as abdominal pain and ischemia, cases have been reported involving arteries throughout the body, including coronary arteries. Compared to SCAD, SAM is typically seen in older patients. There can be considerable clinical and radiographic overlap between SAM and other nonatherosclerotic causes of coronary artery dissection. Histology remains the gold standard for definitively distinguishing between these conditions, a distinction which can have important therapeutic implications.

Sudden Cardiac Death, Coronary Artery Dissection, Segmental Arterial Mediolytic

G46 Characterization of Cadaver Decomposition Islands Using Necrophilous Insect Diversity and Soil Metagenomic Analyses

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After attending this presentation, attendees will gain insight on vertebrate decay, insect scavenging, the effects of cadaver decomposition islands on topsoil microbial community structure, and the application of metagenomics in forensic science.

This presentation will impact the forensic science community by demonstrating the importance of understanding

biological and ecological aspects of the carrion community.

Introduction: Decomposition of large vertebrates above ground is predominately driven by microorganisms and necrophilous insects. Scavenging activities of sarcosaprophagous insects dramatically alter Cadaver Decomposition Islands (CDIs) both mechanically and chemically, resulting in nutrient-loading of decomposition products in topsoils and exposure of internal resources for later-arriving species. In forensic taphonomy, site ecology influences the rate of decay while decomposing remains affect a site's microhabitat. Thus, analysis of soil microbial community structure, in conjunction with necrophilous insect diversity and succession, associated with CDIs is critical to fully understand the carrion habitat. The widespread availability and low-cost of next generation sequencing makes cadaver soil metagenomic analyses feasible and potentially a valuable tool in forensic science.

Materials and methods: Four 12-month long seasonal studies (winter, summer) were conducted in a hardbottom flatwoods forest in Hammond, Louisiana, from February 2011 to July 2013. Each field study included three adult swine carcasses (~45-122kg, with average weight ~68kg) placed directly on the leaf litter/soil surface. Sampling events and protocols varied for insect and soil core collections throughout the five stages of decay. Manual sampling and insect pitfall traps (four traps/carcass) were collected daily, every other day, biweekly, bimonthly, and monthly. Two soil cores (~12cc each) were collected beneath each carcass every 3 days, biweekly, bimonthly, and monthly. Control soils were collected per sampling event ~15m away from the carcasses.

Soil samples were processed for soil characterization and chemical composition (pH, KCl extractable organic carbon, total nitrogen, ammonia/ammonium, orthophosphate, and nitrite). Microbial characterization included enumeration of lipolytic and proteolytic colony-forming units (three serial dilutions in triplicate/sampling event) and 16S rDNA metagenomic analysis using semiconductor sequencing (Ion Torrent PGM™ with barcoded universal primers flanking the V4 region). Statistical analyses of insect and microbial data included classification of insects and amplicons, community structure (α/β diversity), and succession analysis and ordination using principle component analyses.

Results: A total of 35 and 34 soil sampling events (280 and 272 total soil cores, respectively) were performed during the winter and summer 2011 series, respectively. *Calliphoridae* and *Sarcophagidae* immatures were the predominant insect scavengers of swine carrion during the fresh to active decay stages for both winter and summer studies, including the following blow fly species: winter 2011-2012 studies: *Calliphora vicina* (Robineau-Desvoidy), *Phormia regina* (Meigen), and *Lucilia coeruleiviridis* Macquart (*Calliphoridae*); and summer 2011-2012 studies: *Cochylimyia macellaria* (F.), *Chrysomya rufifacies* (Macquart), and *L. Coeruleiviridis* (*Calliphoridae*). Later stages of decay were primarily associated with the following fly species: *Hydrotaea leucostoma* (Muscidae) and *Fannia scalaris* (F.) during active to putrid remains; and *Hermetia illucens* (*Stratiomyidae*) from advanced decay to putrid/dry remains.

Shifts in soil microbial biomass reflect, in part, the nutrient-loading of soils with cadaveric fluids due to necrophagous insect activity. For instance, increased lipolytic activity was observed from early advanced decay to late putrid/dry remains stages for both winter and summer studies (e.g., winter 2011: ~days 35-200; summer 2012: ~days 12-180 of decay). Trends in proteolytic microbial biomass were similar for control and cadaver soils during fresh, bloat, active, and late putrid/dry remains stages. Thus, increased proteolytic activity was observed during ~days 40-200 and ~days 57-130 of decay for winter 2011 and summer 2012 studies, respectively.

Family level shifts in microbial diversity correlated with the five stages of decay. For example, the winter 2011 soils demonstrated

the following trends: (1) control, fresh, and bloat stages were dominated by *Acidobacteria*; (2) microbes present during active and advanced decay were predominately *Pseudomonadaceae* and *Flavobacteriaceae*; and, (3) putrid/dry remains were dominated by *Xanthomonadaceae* and *Chitinophagaceae*. Clustering of stages of decay based on microbial and insect data were observed using principle component analyses.

These results suggest that stages of decay could potentially be characterized using microbial topsoil community composition. From an evidentiary point of view, the more information one has, the stronger the case. Thus, investigating the biotic and abiotic aspects of CDIs is imperative to our understanding decomposition.

Decomposition, Forensic Entomology, Soil Metagenomics

G47 Accidental Opioid-Induced Deaths: Modeling Relationships of Postmortem Opioid Levels to Co-Intoxicant Benzodiazepine, Alcohol Presence, and Selected Decedent Characteristics

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After attending this presentation, attendees will better understand the potential impact of co-intoxicant benzodiazepines, alcohol, and other decedent characteristics in opioid-induced deaths involving oxycodone, methadone, hydrocodone, or fentanyl.

This presentation will impact the forensic science community by providing research which can help medical examiners sort out toxicological complexity in opioid multi-drug deaths, which have increased in frequency.

Accidental poisoning is the second-highest cause of accidental death nationally, surpassing motor vehicle fatalities. Rural states, including the study population, have experienced unexpected increases in such deaths, which involve both prescribed and diverted drugs. The four most common opioids found in toxicology tend to be oxycodone, methadone, hydrocodone, and fentanyl, and the two most common benzodiazepines tend to be alprazolam or diazepam. Alcohol is also frequently involved. Despite the frequency of polydrug deaths with these drugs found in toxicology, there has been little research that addresses the complex relationships among them.

A Forensic Drug Database (FDD) was created to capture drug death data. A project funded as part of the West Virginia Injury Control Research Center's renewal grant from the Centers for Disease Control and Prevention (CDC) expands the FDD from West Virginia (WV) to the northern New England (NNE) states of Maine, New Hampshire, and Vermont. Decedent data collected in the medical examiner files is entered into the FDD, including demographic information, body condition, Body Mass Index (BMI), death certificate data, route of drug administration, whether a prescription was present for controlled substances identified, medical history, key autopsy findings, and toxicological analyses. The database utilized here contains information on WV drug-related deaths from January 2005 through most of 2010; data from

NNE are being compiled.

Opioid-induced deaths in West Virginia were analyzed using medical examiner case files and a comprehensive forensic drug death database. This analysis is part of a larger study comparing opioid mortality and co-intoxication in four rural states. The focus of this presentation is on relationships between opioid levels and the presence of benzodiazepines and alcohol in West Virginia's decedent toxicology findings.

Due to significant deviations from the normal distribution in the opioid concentrations, natural logarithmic transformations were used, resulting in primary analysis of log-concentrations of each opioid. In order to examine associations between particular covariates of interest (age, BMI, gender, benzodiazepine presence, and alcohol presence) and opioid concentrations, multiple linear regression models on the log-concentrations were employed. Since the goal was to build the most parsimonious model (i.e., identify which covariates best predict opioid concentrations), backward model selection methods with an inclusion criterion of 0.1 were utilized.

Out of a total of 2,355 accidental drug deaths, 877 met the criteria for inclusion. These deaths had oxycodone, methadone, hydrocodone, or fentanyl as the only opioid present, with or without alprazolam or diazepam (but no other benzodiazepine), and with or without alcohol. This dataset included 135 deaths involving fentanyl, 135 involving hydrocodone, 337 involving methadone, and 270 involving oxycodone. Only those deaths with femoral or subclavian specimens were included.

The following significant relationships were found, accounting for covariates. Among deaths caused by oxycodone, alcohol is significantly associated with a decrease in the log-concentration of postmortem levels of oxycodone ($p=0.0011$). Among deaths caused by methadone, alcohol is significantly associated with a decrease ($p=0.0002$), benzodiazepine presence with a decrease ($p=0.0153$), and increasing age with an increase ($p=0.0420$) in the log-concentration of postmortem levels of methadone. Among deaths caused by hydrocodone, alcohol is significantly associated with a decrease ($p=0.0329$) and benzodiazepine presence with a decrease ($p=0.0547$) in the log-concentration of hydrocodone. Among fentanyl deaths, there were no significant relationships observed ($p=0.77$).

In conclusion, the four opioids in the model responded differently to the presence of alcohol, specific benzodiazepines, and age. Alcohol presence was significantly associated with decreased log-concentrations of oxycodone, methadone, and hydrocodone. Benzodiazepine presence was significantly associated with decreased log-concentrations of methadone and hydrocodone. Increasing age was only associated with an increase in one opioid, methadone. None of the variables were significantly associated with postmortem concentrations of fentanyl. BMI and gender were not significantly associated with concentrations of any of the four opioids.

Accidental Poisoning, Opioid, Postmortem Toxicology

G48 Retinal Hemorrhage and Axonal Injury in Pediatric Traumatic Deaths

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After attending this presentation, attendees will be able to identify the association between retinal hemorrhage, axonal injury, and the possible mechanisms of traumatic injury in the pediatric population.

This presentation will impact the forensic science community by discussing the relationship of retinal hemorrhage, axonal injury, and traumatic injuries in the pediatric population.

The right and left globes were removed from the 42 pediatric-age decedents from three forensic pathology centers and referred for examination as part of a work-up for cause and manner of death. The specimens were placed in neutral buffered formalin for a minimum of 48 hours without pre-injection. After examining each specimen externally for optic nerve sheath hemorrhage and serially sectioning the optic nerve for histology, the globes were oriented by identification of the inferior oblique attachment and sectioned in the horizontal plane, to include optic nerve and globe. Sections were dehydrated in serial ethanol and xylene solutions, embedded in paraffin, and sectioned at 8 microns. Slides were deparaffinized and stained with Hematoxylin-Eosin (H&E) and Prussian blue. Immunohistochemistry using antibodies against amyloid-beta precursor protein (monoclonal antibody clone 22C11) according to an established protocol, along with positive and negative controls, was performed on all cases. The prepared slides were evaluated blindly by a neuropathologist. Trauma history was obtained from the investigative and autopsy reports.

A subset of cases with investigation- and autopsy-confirmed abusive injuries had evidence of retinal hemorrhage, as expected. All cases showed intraretinal hemorrhage with a tendency for small hemorrhages within the nerve fiber layer between the internal limiting membrane and ganglion cell layer, although other layers were involved as well. They tended to occur throughout the retina and extend anteriorly to, or near, the ora serrata. Some cases also showed preretinal, subhyaloid, and/or vitreous hemorrhage in addition to the intraretinal hemorrhage. Prussian blue staining for iron demonstrated iron deposits (remote hemorrhage) within the dura of the optic nerve as well as focally within colloid in an occasional case. Interestingly, review of the specific investigation findings suggested repeated abuse accompanied by shaking. Also interestingly, immunostains for amyloid precursor protein showed no evidence of axonal trauma involving the retina or optic nerve. In summary, these findings indicate that diffuse traumatic axonal injury involving the retina and optic nerve occurs infrequently, if at all, in abusive head injury in children or in otherwise abusive infant deaths, raising questions about presumed mechanisms of retinal hemorrhages. The occasional case with remote hemorrhage suggests a role for Prussian blue staining to substantiate repeated abuse, although it is likely not a sensitive marker in this circumstance.

Retinal Hemorrhage, Amyloid Protein, Trauma

G49 Wrist Fracture Resulting From an Accidental Electric Shock in a 6-Year-Old Girl

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After attending this presentation, attendees will be aware that low-voltage current exposure may cause radius fractures in children, due to a tetanic contraction of the forearm muscles.

This presentation will impact the forensic science community by raising awareness of the need of a radiographic evaluation in a victim who has been exposed to a low-voltage electric shock and who's complaining of a bony or a joint pain, even in the absence of a direct blunt trauma or a fall.

A 6-year-old girl was admitted to the emergency room with right wrist pain about one hour after sustaining an electric shock. She reported getting a shock associated with a burning sensation as she touched, with her right hand, a street lamp supplied by a 230V alternating current, while she was holding onto a metal bicycle rack with her left hand. Both hands were wet. Her father, who witnessed the scene, said she had managed to pull her hands free within seconds of the shock and that she had not fallen or lost consciousness. A few minutes after the accident, the child began to complain of pain in her right distal forearm.

The patient was evaluated by a pediatric physician and an orthopedic surgeon. She was conscious and had normal vital signs. Small, whitish, round and indurated skin lesions with a darker central depression were noted on the palmar aspect of the fingers of her right hand. They were consistent with electrical burns, and especially with entry points of the current, also known as "electric marks of Jellinek." Examination also revealed a right swollen and tender wrist with a normal range of movement. There was no open wound or bruising of the wrist. No anomaly was found after a complete physical exam, including a careful examination of the rest of her skin and extremities, looking for an exit point of the current.

The electrocardiogram and the biological check-up, including cardiac enzymes, were normal. However, X-rays showed an incomplete fracture of the distal metaphysis of the right radius with an anterior displacement. A circular forearm plaster cast was applied for three weeks. The girl was monitored and observed for 24 hours in the hospital before being discharged.

Because bones have high resistance compared to surrounding tissues, fractures are usually seen with high-voltage current exposure (over 1000V) or with additional traumas, such as falls. However, some cases of fractures resulting from electric shocks without any direct trauma and involving low-voltages have been reported in the literature. They usually involve bones of the proximal appendicular skeleton like the scapula, the proximal humerus (sometimes associated with a posterior luxation), or the femoral neck, which are surrounded by large muscle bulk. Vertebral fractures have also been described. These fractures occur as a result of a violent and involuntary muscle contraction. In this case, the torus (or buckle) fracture was caused by a spasm of the forearm flexor muscle group.

Interestingly, only four cases of radius fracture resulting from accidental electric shock have previously been reported in the literature, all of them involving pediatric patients.¹⁻⁴ This case is the youngest ever reported. Children's vulnerability to this type of fracture may be explained by low bone strength close to the epiphyseal growth plate.

Even if rare, the possibility of a radius fracture should always be considered and a radiographic evaluation be recommended in a child who has been exposed to a low-voltage electric shock and who's complaining of a pain in his forearm, even in the absence of a fall.

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Electric Shock, Radius Fracture, Muscle Contraction

G50 Trends in Pediatric Suicides in Chicago From 1993 to 2013

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After attending this presentation, attendees will have a better understanding of the incidence and circumstances surrounding pediatric suicide in a busy, urban medical examiner's office during the last 20 years.

This presentation will impact the forensic science community by providing an overview of pediatric suicides in Chicago, which may be useful in determining risk factors and/or prevention strategies for the pediatric population.

According to the Centers for Disease Control and Prevention (CDC), suicide is the third leading cause of death in young people between the ages of 10-24 years old. It is a serious public health concern that is often overlooked. A search of the records of the Cook County Medical Examiner's Office over the past 20 years (1993-2013) identified 285 suicides that occurred in subjects who were age 17 years old and under. The youngest was 9 years old. The mean age was 15.1 years of age and the median age was 16 years old. The cohort was then divided into elementary school age (<11 years old), middle school age (11-13 years old), and high school age (14-17 years old). Among the cohorts studied, the suicides among elementary school-age subjects were 2.8% (n=8). In middle school-age subjects, the occurrence was higher at 15.1% (n=43). High school-age subjects showed the highest proportion of suicide at 82.1% (n=234). The leading cause of suicide in this cohort was hanging (46.7%; n=133), followed by gunshot wounds (38.9%; n=111), and trains striking pedestrians (4.9%; n=14). Single and/or combined drug intoxication and carbon monoxide intoxication via automobile tied for the fourth leading cause of suicide in the pediatric population (1.8%; n=5). The remaining causes of suicide among this cohort included falls from heights (1.4%; n=4), shotgun wounds (1.1%; n=3), drowning (n=2), incised wounds (n=2), automobiles striking pedestrians (n=2), asphyxia (n=1), motor vehicle collision (n=1), jumping from a vehicle (n=1), and thermal burns (n=1). Consistent with national statistics, there was a male predominance with 209 (73%) of the subjects being male and 76 (27%) female. By race, 199 (69.8%) of the subjects were White, 84 (30.2%) were Black, and 2 (0.01%) were other, both of whom hanged themselves. Over this 20-year period, the incidence of suicide in the pediatric population has, for the most part, remained steady (mean incidence = 14.25 cases per year). Peak incidences occurred in 1994 (n=23) and 1997 (n=21). The years 2005 (n=5) and 2006 (n=3) had the lowest rates of pediatric suicide deaths.

Suicide deaths in the pediatric population are a poorly studied area in forensics. Debates regarding the age at which a child can form intent to commit suicide and the most appropriate way to classify the manner of death in such cases are ongoing. This is the first forensic study of pediatric suicide in the Chicago metropolitan area and the intent is to offer unique information for future intervention programs and a basis for forthcoming research.

Pediatric, Suicide, Chicago

G51 Ganglia and Nerve Root Hemorrhage in Cases of Pediatric Blunt Head Injury

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The goals of this presentation are to introduce a novel method for extraction of the entire spinal cord with the attached dorsal root ganglia and to present the results of a pilot study comparing histological findings of pediatric decedents with and without documented trauma.

This presentation will impact the forensic science community by introducing a novel method for spinal cord extraction that is more cost- and time-efficient than the previously published *en bloc* method as well as a concise method for scoring the presence or absence of nerve root and dorsal root ganglia hemorrhage. A preliminary comparison of hemorrhage in nerve roots and/or ganglia within cervical, thoracic, and lumbar regions between traumatic and non-traumatic deaths will be presented.

Downs, Downs and Alexander, and Matshes *et al.* all have presented preliminary data supporting a relationship between cervical nerve root and ganglion hemorrhage and abusive head trauma.¹⁻³ Downs and Downs and Alexander were the first to describe a method for extracting and examining cervical nerve roots and ganglia and identified hemorrhage in these structures of children with a history of abusive head injury.^{1,2} Using the same extraction technique, Matshes and colleagues found nerve root hemorrhages in the cervical ganglia that they attributed to tensile forces associated with the back-and-forth motion of the neck during shaking.³ The extraction technique used in each study involved removal of the cervical vertebral column with enclosed neurological tissues *en bloc*, followed by histologic preparation of the entire specimen. The method was labor intensive, costly, time consuming, and highly invasive; additionally, histologic processing of the spine required decalcification of the specimen which may also introduce artifacts or interfere with any subsequent immunohistochemical studies.

The Harris County Institute of Forensic Sciences (HCIFS) has developed an alternative method for extracting a spinal cord with attached dorsal root ganglia that is time- and cost-efficient and much less destructive. Extraction of the spinal cord begins with a standard posterior approach. A posterior laminectomy is completed using a reciprocating saw. The lateral column is then removed by cutting through the overlying superior and inferior articulating facets and then the pedicle using Stony Coral Cutters, a manual bone-cutting instrument. The segmented bone is retracted and the underlying ganglion is exposed. Then each nerve is cut lateral to the ganglion with a scalpel. Finally, the complete spinal cord is removed with the attached ganglia. The neurologic tissue is preserved and sectioned following standard methods.

In addition to the time and cost advantage previously mentioned, the proposed method removes only the posterior and lateral regions of the vertebrae, leaving the vertebral bodies articulated; this maintains the structural integrity of the entire vertebral column. Furthermore, the method allows examination of the cervical, thoracic, lumbar, and sacral ganglia without distortion of the body.

To evaluate the extraction method and to obtain preliminary information regarding hemorrhage in all spinal levels,

HCIFS conducted a pilot study. Spinal cords using the proposed method were removed from 26 pediatric decedents. The ages of the decedents at death were 1 to 48 months of age (mean age: 9 months of age). The study population was broken into two groups: a control group with no traumatic findings during the autopsy; and, an experimental group with traumatic findings during the autopsy. The control group included 9 decedents, age range 1 to 6 months. The experimental group included 17 decedents, age range 1 to 48 months. Cranial trauma was identified in 16 decedents of the experimental group. For each decedent, histological sections of nerve roots and dorsal root ganglia were examined for the presence of hemorrhage. Each slide was scored using a binary system of presence/absence. In the control group, hemorrhage was observed in only one (11%) decedent, a documented hemophiliac. In the experimental group, hemorrhage was observed in 16 (77%) of the decedents, all with documented cranial trauma.

Although the pilot study sample population was small, the results indicate that removal of the spinal cord using the proposed method is valid and that hemorrhage in the spinal nerve roots or dorsal root ganglia is an indicator of trauma. HCIFS, in collaboration with a regional pediatric hospital, is conducting a protracted study to more thoroughly investigate the role of nerve root hemorrhage in pediatric deaths.

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Ganglia Hemorrhage, Spinal Cord Extraction, Blunt Head Injury

G52 An Evaluation of the Discriminatory Power of Clinical Diagnostic Findings of Abusive Head Trauma Using U.S. Hospital Data

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After attending this presentation, attendees will understand the degree of diagnostic accuracy of abusive head trauma models for acutely head-injured children.

This presentation will impact the forensic science community by contributing new information to the ongoing medicolegal discussion concerning the clinical basis for a diagnosis of abusive head trauma in children.

The diagnosis of Abusive Head Trauma (AHT) in a child or infant is typically based on the clinical triad of subdural hematoma, retinal hemorrhage, and encephalopathy in an acutely head-injured child without a history of explanatory trauma.¹⁻⁵ The presence of additional findings associated with abuse, e.g., external injuries and bone fractures, *inter alia*, adds strength to the likelihood that the diagnosis is accurate.⁶ The predictive accuracy of the triad and associated findings is unknown, however. There is currently no standard case definition for AHT and no clinical test that distinguishes between intentional and unintentional head injury, and the diagnosis of AHT is therefore made inconsistently, often reflecting personal beliefs, biases, and practices, rather than a consensus-derived gold standard.^{7,8} This absence of a gold

standard will ultimately lead to the over- and under-diagnosis of AHT. The potential for over-diagnosis is particularly of a concern when only triad findings are present and there are no other findings indicative of abuse.⁹

Experimental clinical trials with proper case and control groups are, for obvious reasons, not feasible for an injury such as AHT. In the alternative, other approaches utilizing observational data have recently been employed as a means of evaluating the discriminatory power of diagnostic algorithms for AHT.^{6,10-12} Additionally, the Centers for Disease Control and Prevention (CDC) have developed a standard case definition for non-fatal AHT based on International Classification of Diseases (ICD)- 9 codes for routine surveillance purposes.¹³ The present study assessed the validity of two epidemiologically-derived clinical prediction rules for AHT: the five-variable model proposed by Hymel and colleagues and the six-variable model introduced by Maguire and colleagues.^{6,10} The discriminatory power of both models was validated using U.S. hospital data abstracted from the Kids' Inpatient Database (KID) of the Healthcare Cost and Utilization Project of the Agency for Healthcare Research and Quality for 2000-2009. The KID database contains data from a stratified sample of 80% of pediatric discharges from U.S. hospitals and thus provides users of the database with a large, representative sample size allowing for investigation of rare conditions and treatments, and the extrapolation of the results to the background population. The KID database was queried for children <3 years of age fitting the inclusion criteria of the two models, respectively. Children involved in a traffic accident or whose records were indicated were excluded. Each child was classified as "abused" or "other" based on the presence of an abuse ICD-9 code. The models were then applied by categorizing children with any ICD-9 code corresponding to the variables proposed as indicative of abuse as likely abused and sensitivity, specificity, positive predictive value, and negative predictive value for both models respectively were calculated and compared to the results found in the original studies.

It was found that currently available diagnostic algorithms for AHT have low discriminatory power when present (low specificity and low positive predictive value) but high discriminatory power when absent (negative predictive value >0.9), and thus of limited practical use. Relying solely or too heavily on such algorithms entails a substantial risk of misdiagnosis. Caution in interpreting clinical diagnostic findings absent collateral evidence of abuse is necessary in the investigation of AHT, and the use of triad findings alone as an indication of AHT is improper and potentially harmful.

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Abusive Head Trauma, Forensic Epidemiology, Diagnostic Test

G53 Fatal Accidental Methadone Intoxication in a Child: An Analytical Method to Define the Time of Ingestion

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The goal of this presentation is to show a case of fatal accidental methadone intoxication in a 1-year-old male infant in which the toxicological investigations have helped to define not only the cause of death but also the time of the ingestion of the toxic substance. Postmortem findings with a complete histological and toxicological analysis of methadone distribution in fluids and tissues are discussed.

This presentation will impact the forensic science community by showing the importance of an accurate anamnestic, circumstantial, histological, and toxicological investigation in all drug-related deaths in order to clarify the exact mechanism of death and the time of ingestion of the toxic substance.

Methadone is a synthetic opioid. It was initially synthesized as a morphine substitute in Germany during World War II and approved by the U.S. Food and Drug Administration (FDA) in 1947 for use as an analgesic. By 1950, physicians prescribed it for the treatment of withdrawal symptoms associated with heroin and other opioids. Recreationally, methadone is abused for its sedative

and analgesic effects.

The primary effects of methadone include drowsiness, sedation, dizziness, mood swings (euphoria to dysphoria), depressed reflexes, altered sensory perception, stupor, and coma. Other physiological effects include strong analgesia, headache, dry mouth, facial flushing, sweating, nausea and vomiting, respiratory depression, muscle flaccidity, pupil constriction (miosis), and decreased heart rate. The principal mechanisms by which methadone causes death are discussed: respiratory depression; aspiration of vomit; pulmonary edema; bronchopneumonia; cardiac problems; and, renal failure.

The drug is distributed as a watery solution in very small bottles, which are fitted with an ordinary screw cap. This kind of distribution may have fatal consequences, in particular for children. The literature shows numerous fatal accidental methadone intoxications in children. In these cases, postmortem (external and internal) examination and toxicological analyses are indispensable for the final clarification of the cause of death but also, as this case demonstrates, to establish the time of ingestion of the substance.

A 1-year-old male infant was seen in the emergency room after accidental methadone ingestion. He presented with coma, miosis, non-reactive pupils, respiratory depression, and asystole. The child's father reported to the police that he and his wife were being treated with methadone for heroin addiction. In the afternoon, after taking their usual dose of methadone, they left the bottle, which still contained methadone, on a chest of drawers. Suddenly, they realized that their son had the bottle in his hand and was drinking. Immediately the father took the bottle from the hand of his son and, seeing the baby sleepy and sweaty, put him on the bed. After two hours, he touched the child and realized he was cold and unconscious. When they arrived in the emergency room, the baby was already dead.

A complete autopsy was performed 24 hours after death. The external examination did not reveal any traumatic lesions. The internal examination revealed only polivisceral stasis and massive pulmonary and cerebral edema. Histological examination revealed cytotoxic and vasogenic edema in the brain, endoalveolar edema, bronchial aspiration of food material in the lungs, sinusoidal stasis, microvesicular steatosis in the liver, and subcapsular hemorrhages in the spleen.

Methadone was detected in the subject's urine through immunoenzymatic screening. Toxicological analysis by solid-liquid extraction and Gas Chromatography/Mass Spectrometry (GC/MS) analysis was carried out to identify and quantify the lethal substances present in the standard biological fluids and organs. Concentration of methadone was determined as follows: blood 1.06mcg/ml; bile 1.26mcg/ml; urine 0.53mcg/ml; brain 0.19mcg/ml; liver 3.89mcg/ml; and kidney 0.96mcg/ml. 2-ethylidene-1,5-dimethyl-3,3-diphenylpyrrolidine (EDDP) concentration was: blood 0.34mcg/ml; bile 3.56mcg/ml; urine 0.56mcg/ml; brain 0.06mcg/ml; liver 1.66mcg/ml; and, kidney 0.63mcg/ml.

To define the time of the accidental ingestion of the substance, a toxicological analyses was also performed on the gastrointestinal tract. In these samples, methadone concentration was: gastric contents 19.88mcg/ml; duodenum 0.29mcg/ml; ileum 0.37mcg/ml; colon 0.36mcg/ml. EDDP concentration was: gastric contents 6.99mcg/ml; duodenum 0.33mcg/ml; ileum 0.3 mcg/ml; and colon 0.13mcg/ml.

Chromatographic investigation was also carried out on the hair of the child. It revealed traces of methadone which are the result of environmental contamination and not due to the intake of the substance. All other substances of abuse and alcohol were negative.

According to the crime scene data, autopsy, and

histological and toxicological findings, death was attributed to an acute intoxication by accidental ingestion of methadone, which occurred three to five hours before the death of the child, certified upon arrival at the emergency room.

Methadone Intoxication, Time of Ingestion, Toxicological Findings

G54 Congenital Cystic Adenomatoid Malformation (CCAM): A Rare Case of Sudden Infant Death

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The goal of this presentation is to examine the histopathological aspects of a case of type II Congenital Cystic Adenomatoid Malformation (CCAM).

This presentation will impact the forensic science community by describing the necessity for a complete methodological forensic approach by means of autopsy and histopathological examinations to diagnose CCAM, often detected before birth by a routine ultrasound scan.

CCAM of the lung is an uncommon cause of respiratory distress in neonates and babies due to a developmental anomaly of the terminal respiratory structures. The incidence of CCAM is 1 in 25,000-35,000 pregnancies.¹ This disorder can be diagnosed in the neonatal period, but up to 90% of diagnoses are made within the first two years of life.^{1,2} It has been described in association with bronchopulmonary sequestration, extralobar intra-abdominal sequestration, or bronchial atresia in live and stillborn babies. CCAM may be associated with polyhydramnios. CCAMs are classified into three different types based largely on their gross appearance. Type I has large multiloculated cysts (>2cm) and accounts for 50%-70% of cases.³ Type II is composed of smaller, uniform, evenly spaced cysts that rarely exceed 1.2cm in diameter. Type III is not grossly cystic and is referred to as the "adenomatoid" type. Microscopically, the lesions are not true cysts, but communicate with the surrounding parenchyma. The cysts are lined with ciliated cuboidal to columnar epithelium overlying a fibromuscular layer. Rare cases show malignant transformation, usually to Bronchioloalveolar Carcinoma (BAC), although sarcomatous and blastomatous transformations have also been reported.⁴ It is rarely encountered in adults. This presentation reports on a case of type II CCAM in a 38-day-old female who suddenly died in her crib. This study describes the clinical features, with radiological and histopathological findings.

A 38-day-old infant was found dead in the crib by her father. The body displayed no external injuries or other abnormalities. The radiological study with chest X-ray showed a massive pneumothorax of the left thorax. At autopsy, all the thoracic organs were in *situs solitus* and no fluid was found in the pleural spaces. The autopsy was carried out according to the Letulle technique, removing all organs *en bloc* and conducting the macroscopic examination after fixation in buffered formalin. The lungs showed many emphysematous air bubbles on the pleural

surfaces with diffuse crackling (crepitus) to the touch. The gross section of the lungs showed a brownish and diffusely porous parenchyma because of the many air bubbles which measured up to 0.4cm in diameter. The etiopathogenetic definition was outlined by histological examinations performed on lung tissue samples using Hematoxylin-Eosin (H&E) which revealed the presence of focal pleural fibrosis, large pleural air bubbles bounded by thickened septae, and of atelectasic and collapsed parenchyma alternating with areas of acute and chronic emphysema, characterized by alveolar cavities. An immunohistochemical staining method was also performed in order to qualify the adenomatoid aspects of the cysts. Histological sections of the remaining organs were unremarkable. The death was attributed to acute respiratory failure caused by the CCAM-induced left spontaneous pneumothorax.

In conclusion, death may occur in up to 30% of all affected babies who present with CCAM soon after birth. Risk factors for a poor outcome include other associated abnormalities, microcystic CCAM (type II), and a large lesion in the parenchyma of one or both lungs. A complete methodological forensic approach by means of autopsy and histopathological examination to diagnose CCAM plays an irreplaceable role in determining the exact cause of death.

Congenital Cystic Adenomatoid, Neonatal Death, Lung Disease

G55 Fatal Milk-Induced Pulmonary Hemosiderosis (Heiner Syndrome) in an Infant

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After attending this presentation, attendees will understand that sudden unexpected death in infancy can result from pulmonary hemosiderosis caused by milk protein intolerance.

This presentation will impact the forensic science community by stressing the importance of examining infant lung tissues stained with iron stains to aid in the diagnosis of pulmonary hemosiderosis and to highlight the association between pulmonary hemosiderosis and milk protein intolerance. To date, there have been no known previous reports in the medical literature of sudden death in an infant due to pulmonary hemosiderosis associated with milk protein intolerance attributed to Heiner syndrome.

Sudden unexpected deaths in infants often pose a unique and sometimes complex set of challenges for forensic investigators. A thorough scene investigation, complete autopsy, and review of the circumstances of death and clinical history are required before rendering an accurate opinion of the cause of the infant's death. The Cuyahoga County Medical Examiner's Office routinely stains lung tissues of infants with a Gomori iron stain to better highlight the presence or absence of pulmonary hemosiderosis. The differential diagnosis for the etiology of pulmonary hemosiderosis includes both trauma and natural processes. The reported case presentation will highlight a rare cause of pulmonary hemosiderosis associated with an infant with intolerance for milk protein.

Heiner syndrome is a food hypersensitivity pulmonary disease that affects primarily infants and is most commonly caused by cow's milk. Only a few reports have been published in the medical literature, which may be due to its clinical misdiagnosis. It has been reported that infants with Heiner syndrome that are fed cow's milk from birth will develop chronic respiratory symptoms beginning between the first and ninth month of age. Respiratory symptoms include cough, wheezing, hemoptysis, nasal congestion, and dyspnea. Other symptoms may include recurrent otitis media, recurrent fever, anorexia, vomiting, colic, diarrhea, hematochezia,

and failure to thrive. The clinical diagnosis is supported with a positive milk precipitin test and symptomatic improvement on a trial of milk elimination. Severe cases may be complicated with pulmonary hemosiderosis.

A case is presented of a 3-month-27-day-old male who was diagnosed with milk protein intolerance at 2 months of age. He had a previous medical history including loose stools, persistent nasal congestion, upper respiratory tract viral infection, bilateral conjunctivitis, and poor weight gain. The infant was found by his father, not breathing and unresponsive, in a car seat three hours after the infant was placed in the car seat for a nap. Resuscitation attempts were unsuccessful.

At autopsy, the gross appearance of the lungs included pleural petechial hemorrhages, hemorrhagic pulmonary edema, and dark red-brown parenchymal congestion. Microscopically, the lungs had diffuse interstitial loose lymphoid aggregates, air space edema, and prominent alveolar siderophages. Also, a lymphocytic tracheitis, right otitis media, and changes of congestive heart failure were diagnosed. The cause of death in this case was pulmonary hemosiderosis due to milk protein intolerance, consistent with Heiner syndrome.

Forensic pathologists should be aware of the association between pulmonary hemosiderosis and milk protein intolerance when interpreting potential etiologies of siderophages in infant lungs.

Heiner Syndrome, Milk Protein Intolerance, Pulmonary Hemosiderosis

G56 Investigating a Death Related to Left Ventricular Assist Device (LVAD)

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After attending this presentation, attendees will have an understanding about the process involved in the evaluation of a decedent with an implanted LVAD.

This presentation will impact the forensic science community by raising awareness about an option that has been recently made available for the treatment of Congestive Heart Failure (CHF), the implantable LVAD, of which there are now a variety of devices in developmental stages, in clinical trials, and in routine use. The process of evaluation done at the Regional Medical Examiner's Office in Newark, New Jersey, for a decedent with an LVAD, is described. The CHF scoring, criteria for the use of LVADs, a brief review of the types and functioning of these devices, and their therapeutic complications will be outlined.

A 57-year-old man was transported from his residence to the nearest health care center after being found unresponsive by his spouse. His downtime was estimated at 20 minutes. His past medical history was significant for end-stage CHF, status post LVAD implantation, atrial fibrillation status post pacemaker implantation, gout, hypothyroidism, and obesity (Body Mass Index (BMI): 30.7kg/m²). He was later transported to the major medical center, where his LVAD had been previously implanted. He was diagnosed with anoxic encephalopathy and pronounced dead after failure of resuscitation. At autopsy, he had a 942-gram heart with concentric biventricular myocardial hypertrophy. Gross examination of the heart revealed prominent subendocardial fibrosis in the left ventricular outflow tract and partial fusion of the aortic valve cusps, a finding that has been reported in the literature.¹ Evaluation of

the heart and the device at autopsy revealed no loss of integrity of the anastomosis, no bleeding or evidence of infection. Additional autopsy findings included pleural and peritoneal effusions, passive venous congestion of the liver, and marked pulmonary congestion with numerous heart failure cells. The toxicology screen was negative. Besides the complete autopsy, the LVAD and pacemaker were evaluated by the respective manufacturers.

CHF affects approximately five million patients in the United States and about one-half million new patients are diagnosed every year. While there are pharmaceutical options for the treatment of CHF, the survival rates and quality of life with these therapies remains sub-optimal. An alternative remedy is cardiac transplantation; however, as is well known to the forensic pathology community, the major limitation is the availability of donors for the procedure. The research into the development of an artificial heart was initiated at the National Institute of Health in 1964. In 1994, the Food and Drug Administration (FDA) approved the use of LVADs, initially as a bridge to transplantation. The device is currently used as a destination device for those too ill for cardiac transplantation.

Immediate complications of LVAD use includes post-operative bleeding, including the necessity of massive transfusions with all its associated risks, and infection involving the operative site resulting in sepsis. A slightly more ominous complication is acute gastrointestinal bleeding due to the formation of arteriovenous malformations in the stomach and intestines. Other reported complications include pulmonary insufficiency with right heart failure and transient cerebrovascular ischemic episodes or strokes. The manufacturers are attempting several innovations to reduce these complications.

While there are several articles on the topic of LVAD in the clinical literature, a review of the forensic literature revealed only one article on this topic.¹

Reference:

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LVAD, Congestive Heart Failure, Sudden Death

G57 Non-Arteriosclerotic Myocardial Infarction in a 38-Year-Old Female

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After attending this presentation, attendees will be able to develop a differential diagnosis for the underlying etiology of myocardial infarction, particularly as it relates to spontaneous dissection of epicardial coronary arteries.

This presentation will impact the forensic science community by increasing awareness of the recognition of conditions and risk factors for non-arteriosclerotic myocardial infarction, particularly in women, aiding in proper diagnosis and accurate death certification. Additionally, identification of hereditary thrombophilias followed by appropriate referral of family members embodies our public health function.

A 38-year-old female presented to the emergency department two consecutive days with complaints of chest pain and nausea. On second presentation, she sustained a seizure on transfer to a stretcher. ST elevation and elevated troponin were

documented before death. The decedent did not smoke or use drugs recreationally. At autopsy, the heart was 340 grams and showed transmural infarction with perforation of the posterolateral free wall near the apex, associated with an apparent occlusive, long segment thrombus in proximal circumflex coronary artery. Analysis of antemortem blood from the referring hospital was negative for Factor V Leiden and Prothrombin G20210A mutations. Functional assays for antithrombin and Protein C were normal; however, Protein S activity was 3%. Histological examination of the left circumflex artery showed dissection within the media with reactive inflammation within the adventitia.

Although myocardial infarction is twice as common in men, heart disease is the number one cause of death in women in the United States. The risk of myocardial infarctions in women is often underestimated and symptoms often go unrecognized. There are also studies that indicate that females are less likely to survive a myocardial infarction, particularly younger women. Spontaneous Coronary Artery Dissection (SCAD) is a rare cause of myocardial infarction. Since the condition was first described in 1931, fewer than 200 cases have been reported in the medical literature. There are currently no known direct causes of this condition, although some correlations have been noted. Many patients are women (73% of cases) in the peripartum period or of childbearing age (mean age: 39 years), with few or no risk factors for coronary artery disease. Other associations include contraceptive use and connective tissue disorders, Ehlers-Danlos and Marfan syndromes, and autoimmune vasculitides such as polyarteritis nodosa, Churg-Strauss Syndrome, systemic lupus erythematosus, and antiphospholipid syndrome. Most of the reported dissections have occurred in the left anterior descending coronary artery.

The most common presentation is sudden cardiac death (60% to 80%). SCAD also causes angina, myocardial infarction, arrhythmias, and pump failure. To be classified as spontaneous, dissection must occur in the absence of trauma, previous surgery or catheterization, or extension of an aortic dissection. Coronary artery dissection, while having some morphological and mechanistic similarities to aortic dissection, seems to be pathogenetically distinct. Unlike aortic dissections, no association with hypertension has been noted. Many of the proposed pathogenetic mechanisms for SCAD pertain to changes in vascular wall properties leading to weakening of the media. These include changes in smooth-muscle-cell metabolism, the effect of proteases released from eosinophilic infiltrates, and pregnancy-related changes in connective tissue. The striking predilection for women and histopathological findings suggest changes in the arterial wall due to hormonal influences.

Protein S serves as a cofactor for Protein C to inactivate factor Va and factor VIIIa in the coagulation cascade. Low Protein S activity is associated with acquired conditions such as liver disease, contraceptive and menopausal hormone use, pregnancy, acute and chronic inflammatory illness, sepsis, consumption by acute thrombosis, or concurrent warfarin therapy. Deficiencies of Protein S, Protein C, and antithrombin are associated primarily with an increased risk for venous thrombosis, and seem to play little or no role in development of arterial thrombosis, leading to heart attack and stroke. However, a recent study suggests that Protein C and Protein S deficiencies may be associated with an increased risk for arterial thrombosis in people younger than 55 years of age.

Myocardial Infarction, Coronary Artery Dissection, Hereditary Thrombophilia

G58 Sudden Cardiac Death: Integrating New Technologies Into Forensic Practice

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After attending this presentation, attendees will learn about modern technologies introduced recently in evaluation of Sudden Cardiac Death (SCD) cases, i.e., postmortem genetic testing, also called molecular autopsy, and postmortem radiological examination by Computed Tomography (CT), CT-angiography, and Magnetic Resonance Imaging (MRI). Attendees will better understand the possibilities and limitations about the new role of forensic pathologists considering the genetic origin of pathologies resulting in sudden cardiac death.

This presentation will impact the forensic science community by improving understanding of the advantages and limitations of new technologies introduced in forensic pathology.

The goal of this presentation is to present the current approach for investigating cases of SCD, especially in the young, after the introduction of new technologies into forensic practice. Some practical cases will be presented.

Any sudden, unexpected, or unexplained death is a reason for medicolegal investigation. The underlying cause of sudden death is most frequently cardiovascular, with ischemic heart disease being most common in the general population. Many studies have shown that in victims ≥ 35 years of age, atherosclerotic coronary artery disease is the leading cause of death, whereas in those < 35 years of age, the leading cause of death is sudden unexplained death. Progress made in the fields of molecular biology and human genetics have identified the genetic origin of many cardiac diseases, which can lead to both structural (e.g., hypertrophic cardiomyopathy) and arrhythmogenic abnormalities (e.g., Long QT syndrome, Brugada syndrome) and result in SCD, especially of young victims. Autopsy-negative SCD is most often thought to be the consequence of sudden arrhythmic death syndrome and molecular autopsy is recommended. The potential genetic origin of cardiac pathologies involves the forensic pathologist in the multidisciplinary management of surviving family members. The role of the forensic pathologist is to perform and store the postmortem samples according to legal and ethical guidelines and to direct the relatives of SCD victims to genetic counseling.

Radiological imaging plays a very important role in the diagnosis of cardiac pathologies in the living patient. In recent years, the use of Multidetector Computed Tomography (MDCT), CT-angiography, and cardiac MRI has also been introduced in postmortem investigations of cardiovascular pathologies, mostly related to atherosclerotic coronary artery disease. According to this study's research, the postmortem radiological examination is most often not sufficient to establish the exact cause of cardiac death and at present the autopsy remains the gold standard. More studies are needed in order to validate the postmortem radiological assessment of cardiovascular pathologies, mainly because of the postmortem artifacts, which can mislead the interpreting physician. It is thought by some that in the near future, the combination of postmortem imaging with autopsy will become a new gold standard.

In conclusion, molecular autopsy and postmortem radiology represent very attractive new tools in the evaluation of SCD cases. The role of the forensic pathologist has changed over

time considering that autopsy diagnosis has profound implications for families of the victims and forensic investigations can lead to improvements in the health of the community. The limitations of interpretation of the molecular autopsy and of the radiological postmortem examination should be highlighted, especially in cases involving legal or ethical responsibilities. Over-interpretation of complementary examinations may contribute to an incorrect clinical approach to families of SCD victims and, in some cases, to a miscarriage of justice.

Criteria for performing molecular autopsies and postmortem radiological examinations as well as guidelines for the management and interpretation of postmortem results currently do not exist but should be proposed by teams of international experts. Integration of the new technologies could lead to a better understanding of cardiac pathologies resulting in SCD and, therefore, to a better clinical approach of the living.

Molecular Autopsy, Postmortem Imaging, Sudden Cardiac Death

G59 Early Markers of Myocardial Ischemia Relevant to the Forensic Pathology: An Immunohistochemical and Gene-Expression Study

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After attending this presentation, attendees will get new biological and methodological insights in the field of early myocardial ischemia and will get a panel of highly promising markers to apply to the routine activity in cases of sudden cardiac death.

This presentation will impact the forensic science community by providing solid diagnostic tools for early myocardial ischemia and will also impact human health, as the presented markers can be used to retrospectively analyze some clinical cases, to adjust the existing clinical strategies to prevent sudden cardiac death, and for the estimation of the cardiovascular risk for relatives of the deceased person.

Postmortem diagnosis of acute myocardial ischemia represents a current challenge for forensic pathologists, especially when death occurs within a short period of time (minutes to a few hours) after the onset of the ischemic injury.

Recent works have investigated, at the immunohistochemical level, some markers that accumulate in, or leak from, the human cardiomyocyte after the ischemic event. Nevertheless, these markers are not detectable in the very early phase of myocardial ischemia. Besides, the role of cardiomyocyte apoptosis as a diagnostic tool in cases of early myocardial injury has been investigated by Terminal deoxynucleotidyl transferase dUTP Nick End Labeling (TUNEL) assay, which has shown good sensitivity but controversial specificity.

This investigation has tested, under experimental conditions, the diagnostic potential of some immunohistochemical markers, as well as of the TUNEL assay, for the detection of early myocardial ischemia. Among the immunohistochemical markers investigated are: Troponin I and T, myoglobin, fibronectin (total and tissular), C5b-9, connexin 43, Jun B, and tenascin C. The same and additional markers (as HIF-1 alpha, caspases 3, 8, and 9) have been studied at gene-expression level as well, using the NanoString nCounter® gene-expression system.

A rat model of myocardial ischemia (ligation of Left Anterior Descending (LAD) coronary artery) was used. The immunohistochemical and gene-expression investigations were performed on the ischemic myocardium at different time points after LAD ligation, ranging from 5 minutes to 2 weeks. As comparison, hearts from control- and sham-operated groups were investigated by the same methods. The NanoString nCounter® is a novel gene-expression system which allows direct measurements of mRNA expression levels without enzymatic reactions or bias, with a sensitivity coupled with high multiplex capability, and a digital readout.

The earliest expressions following myocardial ischemia were observed for JunB and dephosphorylated connexin 43 (15 minutes) as well as for apoptosis and hypoxia markers (15-30 minutes), followed by total fibronectin (≤ 1 hour), C5b-9 (≤ 1 hour), myoglobin (≤ 1 hour), and troponins I and T (≤ 1 hour). The latest markers, expressed only in the healing phase of myocardial infarction, were tissular fibronectin and tenascin C.

This study identified, by immunohistochemical and gene-expression investigations performed on a pure experimental model of myocardial ischemia, early markers of ischemic injury as JunB, dephosphorylated connexin 43, and apoptosis effectors, expressed as early as 15 minutes after coronary artery ligation in rats. Moreover, this research confirmed the early expression of total fibronectin, C5b-9, myoglobin, and troponins (≤ 1 hour). This study has, therefore, identified a panel of markers to further apply to the routine forensic practice to improve the diagnosis in challenging cases of sudden cardiac death.

Sudden Cardiac Death, Immunohistochemistry, Gene-Expression

G60 Sudden Death in a Professional Soccer Player Due to Left-Dominant Arrhythmogenic Cardiomyopathy

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The goal of this presentation is to present a case of sudden death during a soccer match of a player affected by an undiagnosed Arrhythmogenic Right Ventricular Disease (ARVD) characterized by an atypical localization in the left ventricle.

This presentation will impact the forensic science community by explaining a sudden unexpected death occurring during physical activity, specifically in professional soccer, in a young man affected by a rare Left-Dominant Arrhythmogenic Cardiomyopathy.

Arrhythmogenic cardiomyopathy generally affects the right ventricle (ARVD) and more rarely invades the left ventricle, too. The basis of this cardiomyopathy consists of a replacement of the heart muscle tissue with fibrous and adipose tissue, associated with an inflammatory infiltrate, caused by myocellular death. This pathologic tissue may cause, in unforeseen and infrequent circumstances, the triggering and the protraction of fast cardiac arrhythmias, including ventricular fibrillation, that may lead to cardio-circulatory collapse. This case report describes a sudden arrhythmic cardiac death due to Arrhythmogenic Cardiomyopathy (AC), precipitated by intense physical exertion.

During an Italian championship soccer match, Serie Bwin, a 25-year-old professional soccer player, fell to the ground, lost consciousness, and died. He had no clinical or electrocardiographic manifestation of heart disease based on prior clinical and instrumental examinations (electrocardiogram, exercise electrocardiogram or stress test, and echocardiography) performed by his football organization.

A complete postmortem examination was performed 24 hours after death. The external examination was unremarkable. The internal examination revealed a normal-size heart and macroscopic features compatible with the agonistic activity performed by the player. Sections of the heart, taken using the "short axis" method, showed an area of scarring in the subepicardial region of the left ventricle and the posterior and lateral areas of septum. In the area of scarring, the wall of the left ventricle appeared thin (0.9cm). The scar tissue measured 0.4cm in thickness and 3.5cm in length. The examination of the right side of the septum and right ventricle did not show any macroscopic alterations. The other organs did not show specific alterations, except for intense vascular congestion and generalized edema.

The histological examination of the heart, performed with hematoxylin-eosin, Acid Fuchsin Orange G-stain (AFOG), and Mallory and Azan stains, revealed normal conformation of the tissue and vessels of the myocardium. In the posterior wall of the septum and in the lateral wall of the left ventricle, a replacement of the ventricular myocardium was present with fibrous and adipose tissue, and the remaining myocardiocytes showed different sizes, vacuolization of sarcoplasm, and large and irregular nuclei. In the right ventricle, with preserved structure, there was an area of subendocardial connective reinforcement and limited interstitial fibrosis. Histological findings of the other organs showed severe stasis. The histological examination led to the diagnosis of left-dominant arrhythmogenic cardiomyopathy as the cause of death.

The limited amount and distribution of altered tissue in the left ventricle justified the normality of clinical and instrumental results obtained during the frequent screening examinations performed annually by the player for his soccer team.

The left-dominant arrhythmogenic cardiomyopathy, although of limited size, is recognized as a high arrhythmogenic potential without evident mechanical consequences on the cardiac pump, which explains, in affected athletes, the possibility of a normal competitive performance until the lethal arrhythmic event.

Arrhythmogenic Cardiomyopathy, Sudden Cardiac Death, Soccer Player

G61 Aortic Dissection Due to Cocaine Use: A Case Series

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After attending this presentation, attendees will gain an understanding of the clinicopathology and the importance of aortic dissection due to cocaine use.

This presentation will impact the forensic science community by discussing the epidemiology, pharmacology, and pathophysiology of cocaine-induced aortic dissection. This topic is important for the forensic science community, but statistics on this occurrence are not well documented. Data accumulated by

the Wayne County Medical Examiner's Office on the incidence of cocaine-induced aortic dissection will be reviewed.

Cocaine is a controlled Drug Enforcement Administration (DEA) Schedule II drug, with reliable lab detection. Cocaine is rapidly metabolized to the inactive compounds benzoylecgonine and ecgonine methyl ester. Ecgonine methyl ester is further hydrolyzed to ecgonine. Cocaine can be metabolized to benzoylecgonine even after death and sample collection. Active metabolites include cocaethylene (cocaine-ethanol byproduct) and norcocaine.

Cocaine is a naturally-occurring stimulant that affects the central nervous system by increasing sympathetic nerve activity, thus increasing norepinephrine release. This leads to sensitization of the adrenergic receptors to catecholamines, which increases sympathetic tone. Increased sympathetic tone leads to increased cardiac activity, vasoconstriction, and hypertension. The induction of rapidly transient hypertension causes shear stress on the aorta, which can then disrupt the aortic wall architecture and potentially result in rupture. Aortic dissection is characterized by longitudinal cleavage of the aortic media by a dissecting column of blood and is initiated by transverse tears through the intima and at least halfway through the media of the aortic wall. The sudden and violent severe elevation of blood pressure induced by cocaine and its active metabolites produces increased shear stress on the aorta. Factors known to predispose to aortic dissection include hypertension, arteriosclerosis, and chronic cocaine use that distort the medial architecture and contributes to the development of cystic medial degeneration.

Between 2002 and 2013, the Wayne County Medical Examiner's Office in Detroit, Michigan, investigated 94 cases of aortic dissection. Eighteen (19.1%) cases were due to cocaine use. There was a 15:3 male:female ratio. Of the eighteen decedents, sixteen were Black and two were White. Mean age was 51.9 years (range 31 to 67), mean body mass index was 27.9kg/m² (range 20.6 to 38.2kg/m²), and mean heart weight was 549.4 grams (range 325 to 825 grams). Of the decedents, 72.2% had hypertension and arteriosclerotic cardiovascular disease, 16.7% had hypertension alone, 11.1% had no evidence of hypertension or arteriosclerotic cardiovascular disease, and 100% had a history of drug use. The mean level of cocaine and benzoylecgonine in the peripheral blood was 149.5ng/mL (range 33 to 410ng/mL) and 1,222.4ng/mL (range 52 to 4800ng/mL), respectively. Additionally, 58.8% had ecgonine methyl ester, 52.9% had methylecgonidine, and 23.5% had cocaethylene detected in the peripheral blood. The types of aortic dissection were as follows: 61.1% DeBakey II/Stanford A; 27.8% DeBakey I/Stanford A; and, 11.1% DeBakey III/Stanford B. The sites of rupture were as follows: 72.2% in the proximal ascending arch of the aorta resulting in a hemopericardium; 11.1% in the arch of the aorta and proximal descending arch resulting in hemothoraces/left hemothorax; and, 16.7% confined to the aorta without identifiable rupture site.

This is the first large case series of aortic dissections due to cocaine use conducted at a medical examiner's office.

Cocaine, Aortic Dissection, Toxicology

G62 Forensic Entomology at the Harris County Institute of Forensic Sciences

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After attending this presentation, attendees will have an appreciation for how useful forensic entomology can be in death investigations, from the routine to the unusual, and how it has been implemented in a busy medical examiner's office.

This presentation will impact the forensic science

community by illustrating the utility of insects and of full-time forensic entomology staff in death investigations.

The Harris County Institute of Forensic Sciences has hired the first full-time forensic entomologist within a medical examiner's office in the United States. This position has created the opportunity to have unprecedented access to entomology during death investigations and the ability to make it a regular part of the investigation process. Forensic entomology has largely been a consultant/academic pursuit, but this position provides the opportunity to work closely with forensic death investigators, crime scene investigators, and forensic pathologists to bridge the academic and applied aspects of the science.

The implementation of forensic entomology services has opened up the application of entomological interpretation to any case where decomposition and/or insects are present. From January 14, 2013 through July 29, 2013, there were 37 entomology cases covering natural, accidental, and homicide classified deaths. Of these cases, 20 were scene investigations and 17 collections of insects were made during autopsy. In cases involving insects, 68.4% of the cases were from indoor scenes and 31.6% from outdoor scenes. This large number of indoor scenes has revealed new challenges to the way that entomological specimens are collected and how temperature data are evaluated and collected inside a structure.

As the database of insects associated with human deaths is being built in Harris County, Texas, several trends have become apparent. Flesh flies (*Sarcophagidae*) and scuttle flies (*Phoridae*) predominate in indoor scenes, with moderate decomposition. While outdoors, the blow flies (*Calliphoridae*) are the most numerous as are the predatory clown (*Histeridae*) and rove (*Staphylinidae*) beetles. Carpet beetles (*Dermestidae*) can be found both indoors and outdoors. The diversity of insects collected from these cases has revealed that there are many species that have the potential to be useful in applying forensic entomology to death investigations, but the tools are lacking for reliable species identification and/or temperature-based development data sets.

The close association between death investigations and forensic entomology has highlighted several areas that challenge current practices and opens the door to exploring new methods in the medical examiner setting. This information will be used to refine our methods and best practices for forensic entomology and enhance our understanding of the ecological processes underlying decomposition and insects.

Insects, Death Investigations, Forensic Entomology

G63 Forensic Entomology and Bacteriology for Postmortem Investigations in Romania

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After attending this presentation, attendees will understand some principles regarding the correlation of information provided by bacteria DNA and necrophagous insect taxonomic analyses, having as an objective accurate postmortem interval identification.

This presentation will impact the forensic science community by serving as a means of reference and comparison for future research regarding the identification of postmortem interval by means of bacteria and necrophagous insect dynamics.

Forensic entomology is a location-dependent science, widely developed in the United States and Europe. Throughout the years, this discipline has gained large amplitude in the field of forensic sciences, starting with classical taxonomy, DNA

identification of adult and immature stages of necrophagous insect species, and entomotoxicology. Meanwhile, forensic microbiology represents a recent research direction. The objective of both fields is to determine the postmortem interval, sometimes with specification of manner and place of death.

In Romania, forensic entomology is not recognized as an additional distinct domain of forensic science and the information that could result from entomological expertise is not used by police forces. Given that this type of study is bioregion specific, research on Romanian territory is necessary and justified. Consequently, this research is interested in determining the succession of necrophagous insect species in Romania and in identifying the microbial diversity dynamics within the animal carcasses for defining microbial targets in the process of decomposition, using exposed pigs as an experimental model.

The study focuses on the characterization of necrophagous insect species' chronological succession and their stages of development while inhabiting pig carcasses exposed in an urban natural environment (Bucharest, Romania). The composition of the corresponding bacterial communities inhabiting the carcasses colon and mouth cavities, in correlation with climate condition and decomposition stages were studied. The experiment was monitored for up to 11 months during cold (November-May) and warm (July-October) periods. The baits were not accessible by vertebrate scavengers and all metrological parameters were continuously recorded.

During each experiment, both adult and immature necrophagous insect species were collected and taxonomically identified. Tissue samples were harvested from internal (4-8cm) and external sections of the pigs' colons and mouths on a weekly basis. Total bacteria genomic DNA was extracted from each sample and bacterial 16S-rDNA fragments were amplified by Polymerase Chain Reaction (PCR). Bacteria diversity was investigated by Denaturing Gradient Gel Electrophoresis (DGGE) analysis and sequencing of electrophoresis gel-extracted DNA fragments.

The results showed an accelerated activity of necrophagous insect species during warm periods. Their diversity in succession was high, with captured species of the families *Calliphoridae*, *Muscidae*, *Anthomyiidae*, *Fanniidae*, *Sepsidae*, *Phiophilidae*, *Cleridae*, *Silphidae*, *Staphylinidae*, *Dermestidae*, and *Histeridae*. The appearances of new necrophagous insect species over time and their development stages showed a correlation with the postmortem interval, meteorological parameters, and carcass decomposition stages.

Bacteria 16S-rDNA DGGE analysis revealed the presence of a high species/strains number in the mouth cavity more so than in the colon during the warm period. During the cold period, the number and representation of bacterial species was constant in both cavities. After ten weeks exposure, new bacterial species appeared in the colon cavity, while in the mouth cavity this diversity occurred after six weeks, a phenomenon associated with temperature increase.

A correlation of bacterial dynamics with the decomposition process and necrophagous insect species activity was observed. Both insect and bacterial communities' dynamics were dependent on the seasonal atmospheric conditions.

The information resulting from this taxonomy and molecular study on necrophagous insects' and bacterial communities' dynamics at two locations of exposed pig carcass in an urban Romanian location represents incipient data that can be used for accurately determining the postmortem interval and for establishing forensic entomology as an official tool in criminal investigations in this country.

Entomology, Bacteria, Forensics

G64 Cases of Psychiatric Diseases and Forensic Entomology

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After attending this presentation, attendees will learn novel information about how the preservation techniques (clothes, cosmetic clay, perfumed oils) can affect the body colonization by insects and the PMI_{min} estimation in cases of bodies preserved by family members affected with psychiatric diseases.

This presentation will impact the forensic science community by demonstrating the importance of a correct crime scene reconstruction focusing on the preservation and conservation techniques used to avoid body decomposition while waiting for the resurrection.

The conservation of the body of a dead family member, typically a parent, while waiting for the resurrection or because of fear of the lost is often associated with psychiatric diseases or with religious convictions. This presentation addresses three cases that occurred in Italy, where three sons with psychiatric diseases or depressive syndromes watched over their dead mothers for weeks. In all cases, entomology was the only approach useful for an estimation of the time since death (minimum Postmortem Interval (PMI_{min})).

At the end of August 2011 in Vibo Valentia, Calabria, the dead body of a 92-year-old woman was found on her bed, dressed in pajamas, and covered by several blankets; the window of the bedroom partially open. The body was in a partially mummified state, with the face and the neck completely skeletonized. No sign of injuries was observed during the autopsy. Due to the poor preservation of the tissues and organs, the cause of death was difficult to establish despite the previous symptoms of cardiac disease. Six species of flies (*Lucilia sericata*, *Chrysomya albiceps*, *Hydrotaea capensis*, *Sarcophaga sp.*, *Piophilidae casei*, and *Megaselia scalaris*), four species of beetles (*Necrobia ruficollis*, *Dermestes frishii*, *Dermestes undulates*, and *Aleochara sp.*), and two parasitic wasps (*Nasonia vitripennis* and *Brachymeria podagrica*) were collected from the recovery scene and during the postmortem examination. The insects' developmental stage correlated with a temperature reconstruction (data logger placed into the bedroom for 5 days) is consistent with an estimated PMI_{min} of 17-23 days. This data is in agreement with the last time the woman had a telephone call from the other son's wife 19 days prior to the discovery of the body.

In April 2012 in Bisceglie, Apulia, a 97-year-old woman was found dead on the floor of her kitchen. Heaters were switched on, thus warming the house to 20°C (68°F). All the windows and entrance doors were sealed. The body was clothed in pajamas and was partially wrapped in several blankets. A low number of active larvae and pupae surrounded the corpse, which was in an advanced state of decomposition and pre-mummification. The face and neck region were covered by a mask of a greenish substance, similar to a powder found in many stoppered glass jars found on the scene.

Additionally, a mixture of essential perfumed oils was abundantly used on the body. A complete autopsy excluded traumatic causes of death and revealed severe ischemic heart disease with 90% coronary artery occlusion. The chemical analysis of the white crystals found on the body revealed that they were coarse table salt granules and the grainy powder was recognized as cosmetic clay. The entomological investigation revealed the presence of two different species of *Diptera*, namely *Calliphora vicina* and *Fannia canicularis*, that allowed a PMI_{min} estimation of 25-30 days.

At the beginning of July 2013 in Livorno, Tuscany, the dead body of an 89-year-old woman was found on her bed, dressed in a nightshirt and nightgown. The window of the bedroom was partially open. Several adult diapers, soaked with decomposition fluids were also present on the bed. The body was lying supine in advanced decay with the face and the forearms partially mummified. No sign of injuries was observed during the external examination of the body and the autopsy revealed severe calcific coronary artery atherosclerosis. Pupae and larvae of *Megaselia scalaris*, *Lucilia sericata*, and *Protophormia terranovae* were collected from the body and the bedroom, allowing a PMI_{min} estimation of 15 days.

The difference in the fauna composition and in the number of species in these cases can be related mainly with accessibility to the bodies (windows partially open vs. windows sealed), the season (summer vs. spring), and the methods utilized in order to preserve the bodies (only blanket and clothes vs. cosmetic clay and perfumed oil).

PMI, Indoor Cases, Forensic Entomology

G65 How Genomics Is Advancing the Field of Forensic Entomology

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After attending this presentation, attendees will have learned how genomic tools are useful, not only to the field of forensic entomology, but to any biological field within the forensic science framework.

This presentation will impact the forensic science community by not only highlighting the latest research which encompasses many biological disciplines such as ecology and evolution, but also aiding in better understanding the basic biological processes that govern the physiological properties forensic entomologists use for criminal justice purposes.

Forensic entomology is a growing field for which there now exists a modest number of individuals who work on this topic within the field of academia. Recently, the first full-time forensic entomologist was hired to work at a medical examiner's office. Because of this advancing field, it is important to develop tools that will aid in death investigations when insects are involved. For example, there are many aspects of forensic entomology that are assumed in the determination of the minimum postmortem interval: effects of abiotic factors such as wind, humidity, and temperature on the oviposition time of eggs; and the effect of biotic factors such as sex and population on the development rate of the insects. Though these factors have been investigated on a basic level (for example, the population structure of some forensically important flies using mitochondrial DNA markers), in this case, a single locus

is investigated and thought to represent the entire genome.

With the availability of whole genome sequencing tools becoming more cost effective, it is now possible to investigate physiological responses to abiotic and biotic factors on a whole genome level; that is, to be able to look to the intersection of individual-level, population-level, and species-level responses. With simple sequencing projects, we are able to isolate and characterize genetic loci that are correlated to the development rate in *Cochliomyia macellaria*. This was accomplished by selecting flies with fast and slow development rates, respectively, and sequencing their entire genomes to isolate genetic markers of development. In the event of a larva to be used in casework analysis, we can then determine the development data that is more appropriate to use given the genotype of the individual. We are able to isolate and characterize genetic loci that are markers of population structure; therefore, in the event of casework, the determination of which appropriate published data set to use can then be more quantitatively determined. And finally, the characterization of the sex of the developing larva can better refine and improve the precision of these minimum postmortem interval estimates.

Forensic Entomology, Genomics, *Calliphoridae*

G66 New Frontiers in Forensic Entomology: Chronobiological Studies on Body Search, Oviposition, and Emergence of *Megaselia scalaris*

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After attending this presentation, attendees will learn novel information about how photoperiod and light-dark conditions can affect forensically important insects in behaviors like oviposition and location of food sources that play an important role for the Postmortem Interval (PMI) estimation.

This presentation will impact the forensic science community by demonstrating how the knowledge of the species chronobiology can improve the PMI estimation, particularly in indoor cases. The presented research deals with *Megaselia scalaris*, a species that plays an important role in colonizing bodies in closed environments.

Circadian clocks have evolved to synchronize physiology, metabolism, and behavior to the 24-hour geophysical cycles of the Earth. The understanding of the circadian clock mechanism is a crucial element of forensic entomology because it is able to control routines such as locomotor activities, locating of food sources, feeding, mating, ovipositing, and emergence times.

Colonization of carrion and human cadavers by insects allow for the PMI to be determined. However, it is thought by some that flies are not active during the nighttime period and, therefore, are not able to oviposit during this time or in general in dark conditions. To put that into a forensic context, if eggs were located on a cadaver, the conclusion would be that death occurred during the previous day or before. Determining nocturnal oviposition in forensically important flies is of fundamental importance so that the PMI can be determined with more precision by the forensic entomologist.

To describe the behavior and the potential role that the circadian clock may have on both the locomotor activity and emergence times of the *M. scalaris*, this study used Trikinetics technology used previously in *Drosophila* studies which allows for factual data rather than observational data as reported in many

articles. The activity rhythms of *M. scalaris* were monitored using light/dark (LD 12:12) photoperiods at 20°C.

Males (N=593, p=0.00) and females (N=205, p=0.00) both demonstrate that there are significant differences in their locomotor activities between dark and light conditions and further results establish that the flies are both diurnal and nocturnal in activity. Both sexes demonstrated a bimodal rhythmical activity which is more evident in the evening. In addition, different light colors have different attractiveness in the two sexes, with females more attracted by red light.

The pupa emergence experiment run in LD 12:12 photoperiod (N=67, p=0.00) determined that there is a significant difference between the emergence in both light and dark conditions, while pupa emergence which was run in complete darkness (N=46, p=0.069) show no significant differences.

To determine if *M. scalaris* were able to oviposit during the nighttime period, 120 females were placed individually into glass tubes containing food at one end and left for 12 hours (7:00 p.m. to 7:00 a.m.). The results demonstrate that, overall, 34.5% of females were able to oviposit during the night.

These experiments have demonstrated that *M. scalaris* is able to oviposit in dark conditions during the night. The pupa emergence determines that there are different rhythms during full darkness conditions and light/dark conditions. In addition, the experiments demonstrated that this species is clock regulated and that in continuous dark it can oviposit both during the dark and the light subjective phase.

The locomotor activity demonstrates that *M. scalaris* is both diurnal and nocturnal in activity which supports the oviposition data also presented.

PMI Estimation, Indoor Cases, Phoridae

G67 *Phormia Regina* and Postmortem Interval Estimations: Wandering Into Curvilinear Blow Fly Development Modeling

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After attending this presentation, attendees will have a better understanding of the importance of forensic entomology in estimating Postmortem Interval (PMI) and the advantages of using curvilinear development data for PMI calculations.

This presentation will impact the forensic science community by vastly improving the current developmental data available for *Phormia regina* and increasing the accuracy of PMI estimations in human death investigations.

The blow fly *P. Regina* belongs to a group of necrophagous insects that can rapidly locate and colonize decomposing animals. When found on a human body, the developing eggs, larvae, or pupae could be used as an index for initial time of death (postmortem interval or PMI) measurements. Estimating the PMI is crucial in most human death investigations, since time of death is needed for proper event reconstruction pre- and post-death. To use insects in estimating PMI, we must be able to determine the insect age at the time of discovery and backtrack to time of oviposition. Consequently, understanding temperature-specific development rates is essential. Unfortunately, existing development models of forensically important insects are only linear approximations.

Here, experiments and findings for building a curvilinear developmental model for *P. Regina* are reported. Experimental considerations include diet, humidity, light cycle, temporal patterns of stage transitions, and temperature measures. Experiments were conducted over 11 temperatures (7.5°C, 10°C, 12.5°C, 15°C,

17.5°C, 20°C, 22.5°C, 25°C, 27.5°C, 30°C, and 32.5°C). Twenty eggs (collected within one hour of oviposition) were placed on 10g of beef liver in a 29.6ml (1oz) plastic cup. The cup was placed in a 4"x4"x2" plastic container that had 2.5cm (1in) of sand in the bottom. Measurements were taken at intervals calculated from Accumulated Degree Hours (ADH). Each life stage had five measurement points: at the beginning; one-quarter mark; one-half mark; three-quarter mark; and, the end. Each point was replicated four times, for a total of 20 measurements per life stage. For each sampling period, the cups were pulled from the chambers and the stage of each maggot was documented morphologically through posterior spiracle slits and cephalopharyngeal skeletal development.

The *P. Regina* data illustrate the advantages of curvilinear models in describing development at environmental temperatures near the biological minima and maxima, and the practical significance of curvilinear models over linear approximations. Results here represent the first in a series of larger studies modeling development of key forensically important blow flies of North America.

Blow Fly Development, Decomposition, Human Death Investigation

G68 Spatial Autocorrelation Using Mantel Tests and Bioinformatics for the Classification and Provenance of Soil Samples for Intelligence and Forensic Applications

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After attending this presentation, attendees will understand that the metagenomic information contained within soils is relevant to the geographic origin of the soil sample. In addition, using bioinformatic machine-learning algorithms, these data can provide soil databases that can be queried when an unknown sample is collected and can provide approximate geographic location and provenance of the source.

This presentation will impact the forensic science community by greatly expanding the forensic geology discipline and enhancing the probative value of soil samples for intelligence gathering and forensic cases. These data can supplement and enhance the investigative and probative value of soil samples.

Based on the ecological hypothesis that soil type (e.g., the chemical/physical properties) drives which microbes live in a particular soil, it should therefore be possible, in theory, to use soil metagenome profiling to produce a unique biotic pattern and associate it to the collection site.^{1,2} If this is indeed true, then a soil biotic profile from a particular geographical location should be able to be mathematically correlated to that location.³ Subsequently, if shown to be true, these soil microbial assemblages would be able to predict soil provenance and have forensic applications. Therefore, the basic questions become: (1) "Is there a correlation between the genetic profile of a soil's microbial community and the geographic location?"; and, (2) "If so, can these predicted patterns be used for soil provenance for intelligence data and/or have a forensic application?" The first question was addressed using a Mantel test for spatial autocorrelation which tests the assumption that geographically closer samples will be more similar than those further apart. Metagenomic data from four taxa — bacteria, archaea, fungi, and plant — from ≈ 1,900 samples collected from primarily

undisturbed soils in Miami-Dade County, Florida, was used. Spatial autocorrelations of genetic data to geographic locations within soil types, from multiple transects, and subplots within subplots were modeled. The analyses used the geographic location (Global Positioning System (GPS) coordinates) and metagenomic biotic profiles from the four taxa. At all scales and in almost all samples, the biotic profiles were positively correlated and significant ($p < 0.05$) to the source of the soil sample. For the two samples that were not significant yet still positively correlated, the historical satellite data were analyzed and it was noted that one site had been burned three months prior to sampling while the other was a disturbed site at an abandoned nursery. This indicated that when there was a deviation from the correlation, the soil was impacted or disturbed in some fashion. Question 2 is being answered using a suite of unsupervised and supervised bioinformatics algorithms that can better predict soil biotic associations to the soil type and specific location.³⁻⁶ This will then allow for a searchable database of soil biotic profiles that is linked to its provenance and can be used for either intelligence or forensic purposes.

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Spatial Autocorrelation, Soil Forensics, Bioinformatics

G69 Microbes and Decomposition: Foundations in Decomposition Ecology and Forensic Applications

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After attending this presentation, attendees will have a greater appreciation for the role microbes play in decomposition ecology and the forensic application of such information.

This presentation will impact the forensic science community by showing that microorganisms play a significant role in the decomposition of human and human analogue remains. Some aspects of this role are predictable and potential exists

for microorganisms to serve as valuable physical evidence in medicolegal death investigation.

Historically, microbes have been overlooked in terms of their importance in decomposition ecology and their application in estimating the time-since-death of human remains. Much of this has been due to the inability to identify the microorganisms associated with decomposition. Traditional techniques rely on culturing microorganisms in the laboratory, which is highly selective and restrictive to a small component of microbial communities. Furthermore, well-established culture-independent techniques to investigate the whole microbial community, such as the analysis of fatty acids, provide little insight into the taxonomy of microbial communities. As a result, lab experiments are often restricted to investigating microbes that are culturable or identifying changes in microbial community structure without knowledge of the taxa involved.

The significance of microorganisms as physical evidence has increased predominately due to the development of novel technologies allowing for the quantification and description of the whole postmortem microbiome. High-throughput sequencing techniques now allow us to examine microbial communities in their entirety from samples taken from the environment. We can now collect microorganisms from a death scene and identify them using 16S and 18S RNA genes. We have learned that these microorganisms are typically identified to phylum and family level, but it is possible to identify microbial species with a high degree (~97%) of reliability. This advancement means that microbial structure and function as related to the greater ecosystem (i.e., human remains found in nature) can be investigated and potentially modeled, validated, and then used in forensic investigations. It is predicted that these developments will result in a forensic microbiology that will contribute unprecedented insight into death investigation and forensic pathology.

A series of presentations will be given that discuss the roles of microbes in the decomposition process of human and animal remains. Researchers from across the United States have been active within this field for the past five years and are now able to present data that provide novel insight into this process. Topics to be covered will be the following: (1) microbial and insect structure and function associated with humans remains and human analogues; (2) the role of quorum sensing by bacteria in regulating arthropod colonization of human remains; (3) the utilization of swine carcasses as models for decomposition; and, (4) using microbes to estimate a minimum postmortem interval.

Taphonomy, Microbiology, Postmortem Interval

G70 Collecting Microbiological Evidence From Cadavers: Techniques and Implications of Using Bacterial Communities in Decomposition Estimates

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After attending this presentation, attendees will learn how to collect microbiological evidence from decomposing cadavers

in order to use previously under-utilized communities of microbes to predict minimum Postmortem Interval (PMI_{min}) estimates and explore phyla-level taxonomic resolution of the microbiological communities sampled from swine carcasses and a human cadaver.

This presentation will impact the forensic science community by providing an approach on how to collect bacterial samples from remains during fresh, bloat, and active decay decomposition stages.

Insects are commonly used for PMI_{min} estimates; however, the potential to use microbiological (bacteria and fungi) samples for forensics has only now begun to be explored. The goal of this study was to describe how to collect microbiological communities from decomposing vertebrate remains while identifying the bacterial communities using state-of-the-art metagenomic sequencing techniques.

Sampling for microbiological communities requires minimal equipment such as sterile swabs, microcentrifuge tubes, and, most importantly, aseptic technique in the field to the greatest ability of the forensic investigator. It is recommended that samples be collected from the decedent at various locations of the body including the buccal and anal region, if possible. Samples should be stored individually and shipped to an expert at -20°C (on ice; dry ice is not necessary). To validate these sampling methods in the field, three *Sus scrofa L.* carrion were placed in a temperate forest in the midwest United States for 5 days. Bacteria communities were sampled at initial field placement after one, three, and five days of decomposition using sterile swabs. Additionally, a human cadaver located at The Forensic Anthropology Center at Texas State (FACTS) was sampled daily from field placement until five days of decomposition. Total genomic DNA was extracted using a modified chloroform-phenol protocol and the bacterial community structure was determined by modified bacterial tagged-encoded FLX-amplicon pyrosequencing. Over the course of decomposition, *Proteobacteria* was the dominant phylum with *Firmicutes* being the next most abundant of the community for swine carcasses. Rare phyla accounted for less than 0.5% of the total relative abundance across decomposition. There was a significant negative linear relationship of phylum taxon richness over the course of decomposition. Similarly, in human cadavers, the most dominant phyla throughout decomposition were *Proteobacteria* and *Firmicutes*.

These data are important because of their implications in forensic ecology and PMI_{min} estimates. The results provide basic knowledge of how to use bacteria colonizing and utilizing carrion for forensic investigations and enhance the understanding of microbiological interactions occurring on the cadaver. Further, this work provides empirical data that may be useful to improve PMI_{min} estimations made by forensic practitioners and potentially increase the use of microbiological communities as evidence at crime scenes. Documenting and identifying differences in bacterial communities is key to advancing knowledge of the carrion necrobiome and its applicability in forensic science.

Microbiological Communities, Evidence Collection, Necrobiome

G71 Postmortem Microbiology: Culturable Bacteria of the Maggot Mass

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After attending this presentation, attendees will understand that a maggot mass is a reducing environment of high temperature that becomes more acidic over time and selects for bacteria commonly associated with putrefaction.

This presentation will impact the forensic science community by providing a better understanding of the ecology of a maggot mass and corpse decomposition, which will help death investigators and forensic entomologists to establish more reliable reconstructions of death scenes and estimates of estimating postmortem interval.

It has recently been shown that postmortem microorganisms have potential value as physical evidence for estimating postmortem interval and tracing the movement of remains. This potential has yet to be fully realized, because we still have only a rudimentary understanding of postmortem microorganisms and their ecology. For example, it is understood that different locations on a corpse have different bacteria; maggot masses are one conspicuous area of a corpse that has yet to be researched. Understanding the microbiology of the maggot mass will provide investigators with a novel source of investigative information that can be used as stand-alone evidence or as evidence that corroborates forensic entomology. This study proposed to characterize culturable postmortem microbial communities and analyze trends in chemical properties.

The experimental unit was a swine carcass (*Sus scrofa domestica*) placed on the soil surface at the Chaminade University of Honolulu Facility for Forensic Taphonomy within a few hours of death. This facility is a 900m² outdoor facility located in a Tropical Savanna climate on the western slope of the Palolo Valley near Honolulu, Hawaii. The site is approximately 285 feet above sea level. Mean annual precipitation equals approximately 700mm, 70% of which arrives in the autumn and winter (October-March). The vegetation at the site is representative of a tropical savanna ecosystem on Oahu; it is rocky and dominated by pili grass (*Heteropogon contortus*) with night-blooming cereus (*Hylocereus undatus*), shrub aloe (*Aloe arborescens*), and carrion plants (*Stapelia* spp.). Few scavengers are present at the site; only the small Asian mongoose (*Herpestes javanicus*) has been observed.

Decomposition was monitored daily for 312 hours postmortem. Maggot masses were present from 70 to 120 hours postmortem and were swabbed daily with sterile cotton swabs and immediately placed in sterile tubes for transport to the laboratory. The swabs were streaked onto standard nutrient agar and incubated at 22°C. Morphologically unique bacterial colonies were isolated and identified via matrix-assisted laser desorption/ionization time of flight mass spectrometry. In addition to swabbing, the chemistry of the maggot masses was characterized using portable sensors to measure temperature, pH, electrical conductivity, and oxidation-reduction potential. Temperature and relative humidity of the decomposition site was measured at intervals of one hour.

Carcasses decomposed to skeletons within 13 days (312 hours) postmortem. Mean maggot mass temperature ranged from 34°C to 40°C, but differences as great as 10°C were observed between masses. The pH of maggot masses was initially slightly

basic (7.4), but became slightly acidic over time (6.6). Measurements of electrical conductivity appeared to be unreliable as values ranged from 19 μ S to 18,000 μ S with extreme variation in between and no consistency among the three carcasses. The oxidation-reduction potential was consistently between -200 millivolts and -250 millivolts, which represents a highly reducing environment related to methane generation and sulfate reduction. The average temperature for each day of decomposition ranged from 24.4°C to 28.3°C, while the mean relative humidity for each day ranged from 68.6% to 96.4%. The most commonly observed bacteria in the maggot mass included *Proteus mirabilis* and *Providencia stuartii*. Also observed were genera *Bacillus*, *Staphylococcus*, and *Enterococcus*. In general, a decrease in bacterial diversity was observed over time. More detailed analyses will be presented and the results observed in this study are the basis for further research in microbe-insect interactions.

Entomology, Taphonomy, Death Investigation

G72 Just How Filthy Are Maggots? Bacteria Associated With the Blow Fly Sister Species *Lucilia Sericata* and *Lucilia Cuprina* (Diptera: Calliphoridae)

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After attending this presentation, attendees will have a better understanding of the bacteria associated with forensically important flies feeding on decomposing vertebrate tissue, how their communities may change over time in association with the insects, and whether such microbes are trans-generationally or environmentally transmitted.

This presentation will impact the forensic science community by clarifying the mechanisms that influence microbial shifts on decomposing remains associated with forensically important flies, as well as the bacterial groups affected by these flies. This is critical information as current information suggests microbial succession on remains is predictive of the postmortem interval and affected by insects.

Improved biological knowledge of flies from the blow fly (Diptera: Calliphoridae) genus *Lucilia* (Linnaeus), especially *L. Sericata* (Meigen) and *L. Cuprina* (Wiedmann) benefits forensic science endeavors.¹⁻³ Since they can be primary colonizers of carrion, developmental data from these species can be useful for estimating the postmortem interval.⁴⁻⁶ Both species engage in myiasis, transmit pathogens, and are also at the center of numerous lawsuits related to the abuse of dependents, pets, and livestock.⁷⁻¹⁰ Accordingly, knowledge of microbial population associated with these flies can help ameliorate the negative and

promote the beneficial properties of *Lucilia* biology.

In all of the examples listed above, there is a likely microbial role that could be investigated. Insect-microbe interactions are well documented.^{11,12} Given the interest in studies of the properties of blow fly-microbe interactions, especially new research targeted toward predicting a postmortem interval with microbial data, a study was conducted to address the following questions: (1) What microbes are associated with these species?; (2) How similar are the microbe communities associated with each species?; (3) What species are trans-generationally transmitted?; and, (4) What species are horizontally transmitted? To address these questions, this study conducted a survey of bacterial communities associated with these sister species using 16S rDNA 454-pyrosequencing.

The results of this study indicated considerable environmental effects on fly-associated communities. Typically, within-generation communities appear to be similar, with trans-generational bacterial communities appearing to turn over quickly. A comparison of female, new liver, old liver, and larval bacterial communities show that there is a small number of potentially important transmitted and facilitated bacteria. Some of these microbes are known pathogens and others are known to affect fly biology.

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Meta-Genomics, Decomposition, Postmortem Interval

G73 An Initial Assessment of the Structure and Function of the Postmortem Human Microbiome: Forensic Applications

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After attending this presentation, attendees will have a greater appreciation and understanding of the bacterial communities associated with human remains and how this information has the potential to be used to make estimates of a minimum Postmortem Interval (PMI_{min}).

This presentation will impact the forensic science community by providing results of some of the first data on bacterial communities associated with human remains, their postmortem community changes, and variation in such changes among epinecrotic communities taken from different areas of a decomposing body.

There are several methods for using biological information in PMI_{min} estimates; however, bacterial assemblages have only recently been investigated for such purposes.¹ With the development and refinement of high-throughput metagenomic sequencing and cost-effective metabolic assay methods, the potential of using data on bacterial succession on human remains for PMI_{min} estimates is quickly becoming a reality.

Epinecrotic communities have been defined as the microbial (i.e., bacteria and fungi) consortia that develop biofilms on the surfaces (external and internal) of decaying organic matter, including animal carrion and human remains.¹ These communities provide the foundation of the necrobiome foodweb and have been reported to affect blow fly attraction to decomposing remains.²

In order to effectively use such information, a better understanding of the postmortem human epinecrotic microbial community, or the microbiome, will be important for developing models to estimate or predict the PMI_{min}. These communities are known to go through a series of taxon replacement over time, much like plant ecological succession or the succession of insect taxon on decomposing remains, a foundation of forensic entomology. Much like insect succession information that is used in forensic entomology to make estimates of the PMI_{min}, recent studies using swine carcasses have demonstrated that postmortem epinecrotic communities show strong promise for use in forensics. This presentation provides some of the first data on the human postmortem epinecrotic communities, demonstrating the potential for using either the taxonomic structure or metabolic functional descriptions of these communities in estimating the PMI_{min}.

This study evaluates the structure (taxonomic) and function (metabolic profiles) of the postmortem human epinecrotic

communities in a series of on-going studies in Texas and briefly discusses how this information can potentially be used in PMI_{min} estimates. A structure/function approach is taken to describe these communities from the skin, buccal, and anal epinecrotic communities. The taxonomic structure of these communities was described using pyrosequencing methods given in an earlier study by a collaborative research group.¹ Metabolic profiles were described using Biolog EcoPlates™ based on methods described by Garland.³

It was found that there were significant taxonomic changes in the postmortem human epinecrotic communities throughout decomposition among sampling days and between sampling areas, but there was no interaction effect. The communities were dominated by the following phyla: *Proteobacteria*, *Actinobacter*, *Firmicutes*, and *Bacteroidetes*. There was an inverse relationship between the relative abundance of *Proteobacteria* and *Firmicutes*, with the latter representing on average 44% of the total community early in decomposition and changing to 13% later in decomposition. There were similar trends in phylum changes over decomposition for each sampling area. However, the epinecrotic community metabolic profiles were not significantly different among sampling days, among sampling areas of the body, and there was not an interaction effect.

These data are an important contribution to the growing understanding of the human microbiome and how these communities can be used postmortem in a forensic context, and demonstrate the potential of using both the taxonomic and functional profiles of epinecrotic communities in estimating the PMI_{min}. Additional research in this area could lead to a greater understanding of the time interval from death to human remains discovery.

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Microbiome, Microbial Succession, Bacteria

G74 Utility of Bacteria Associated With Human Cadavers in Estimation of Postmortem Interval (PMI)

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After attending this presentation, attendees will gain a better understanding of the role played by bacteria and insects during the human decomposition process and how bacterial succession associated with human cadavers can be used for estimating the human Postmortem Interval (PMI). In addition, attendees will also be updated on recent metagenomics approaches developed for the estimation of bacterial community structure from human cadavers.

This presentation will impact the forensic science community by providing a novel PMI estimation method based on bacterial succession alone. This presentation will also provide detailed information on key bacterial groups whose changes in relative abundance may potentially be modeled for the prediction of time since death.

Like insects, microbes also play a significant role in the carrion decomposition process.^{1,2} Studies using pig carcasses in both terrestrial and aquatic environments have shown significant temporal changes in bacterial community structure, but whether the same is true with human cadavers is not clear.^{1,3} In this study, six human cadavers were placed in the field as part of three trials (two bodies per trial) at the Forensic Anthropology Research Facility (FARF) of Texas State University, San Marcos, Texas. Out of six carcasses, primary arthropods (e.g., blow flies, flesh flies, staphylinids) that colonize human remains had open access to three carcasses, while the remaining three were excluded from these arthropods. This design permitted the investigation of the impact of arthropods on bacterial community structure as the remains decomposed. Sterile cotton swabs were used for collection of microbe samples from buccal, skin, and anal regions of the cadaver every eight hours for five days. DNA was extracted from these swab samples using organic extraction method.¹ Three variable regions (V1-V3) of 16S rRNA gene were amplified from extracted DNA using primer pairs 28F and 519R.⁴ 454-pyrosequencing was performed on amplified Polymerase Chain Reaction (PCR) products using the Bacterial Tag-Encoded FLX Amplicon Pyrosequencing (bTEFAP) method.⁵ Sequencing error was minimized using PyroNoise as implemented in Mothur v. 1.29.^{6,7} Low-quality regions of the sequences were trimmed using the sliding window (50 bp; Q35) option in Mothur v 1.29.⁷ All sequences were checked for chimera formation using Uchime as implemented in Mothur v. 1.29, and using the most abundant sequence as a reference datum.^{7,8} Suspected chimeras were deleted and the remaining sequences were utilized for hierarchical classification, α and β -diversity and richness index estimation, multivariate analyses, and for Analysis of Molecular Variance (AMOVA) using Mothur v. 1.29 and R version 2.15.1.^{7,9} Indicator species analyses were also performed in Mothur v. 1.29 for determination of temporally informative species.⁷

Preliminary results from the first trial suggest that bacterial community structure changes significantly over time, and primary colonizing arthropods play a significant role in bacterial succession on human cadavers. At the phylum level, an inverse relationship exists between relative sequence abundances of *Proteobacteria* (increases with time) and *Firmicutes/Actinobacteria* (decreases with time), and it does not depend on arthropod access or exclusion. However, at the genus level, only human remains that were exposed to insect activity resulted in more accurate estimates of their associated PMI. Significantly different bacterial community structures were observed between 0-3 days and 4-5 days of decomposition. *Acinetobacter*, *Wohlfahrtiimonas*, *Anaerococcus*, *Finegoldia*, and *Ignatzschineria* were the top five indicator genera, whose relative sequence abundances varied with time.

In conclusion, this study provides evidence for the first time that bacteria are a potential forensic indicator for human PMI estimation. There is an influence of primary colonizers on bacterial succession. The data obtained from this research can be used for the development of model-based methods for estimation of human PMI, as was previously done using a porcine model.¹

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Bacteria, Forensics, Postmortem Interval

G75A Preliminary Survey of a Postmortem Skin Microbiome on Oahu

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After attending this presentation, attendees will understand how postmortem skin microbial communities change as a carcass decomposes.

This presentation will impact the forensic science community by demonstrating that the postmortem skin microbial community can change rapidly after death. This change is characterized by a shift from a microbial community that includes aerobic bacteria that respire along with representatives from perimortem environment to a microbial community that is dominated by anaerobic fermentative bacteria typically associated with putrefaction, endospore formation, and acidic habitats.

The experimental unit was a swine carcass (*Sus scrofa domestica*) placed on the soil surface at the Chaminade University of Honolulu Facility for Forensic Taphonomy within one hour of death. This facility is a 900m² outdoor facility located in a Tropical Savanna climate on the western slope of the Palolo Valley near Honolulu, Hawaii. The site is approximately 285 feet above sea level. Mean annual precipitation equals approximately 700mm, 70% of which arrives in the autumn and winter (October-March).

The vegetation at the site is representative of a Tropical Savanna ecosystem on Oahu; it is rocky and dominated by pili grass (*Heteropogon contortus*) with night-blooming cereus (*Hylocereus undatus*), shrub aloe (*Aloe arborescens*), and carrion plants (*Stapelia* spp.). Few scavengers are present at the site; only the small Asian mongoose (*Herpestes javanicus*) has been observed. Decomposition was monitored at intervals of approximately 8 hours for 58 hours postmortem. The head was swabbed at 4, 34, and 58 hours postmortem with sterile cotton swabs. At each of the three time points, the head was swabbed five times to help quantify variability within versus across time points. A total of 30 swabs were immediately placed in sterile tubes and transported to the laboratory. Swabs were then stored at -20°C for three days until they were shipped to the Second Genome in San Bruno, California, for sequencing. Interpretations of microbial community structure were limited to phyla and families that represented at least 2% of the whole microbiome.

By 58 hours postmortem, the carcass was in an advanced stage of decay with maggots migrating to pupariate. Throughout decomposition, the postmortem microbiome was dominated by bacteria from phylum *Firmicutes*. The overall community changed significantly ($P < 0.01$) at each sampling time. The microbiome at four hours postmortem was one that was relatively diverse (4 phyla, 14 families) and included taxa commonly associated with skin, fecal matter, or soil. Microbial diversity was less (3 phyla, 9 families) at 58 hours postmortem and comprised taxa that are typically associated with the skin or putrefaction. In fact, most of the families that represented $\geq 2\%$ of the microbiome at four hours postmortem decreased to an abundance of $< 2\%$ by 58 hours postmortem.

Over the course of decomposition, the abundance of bacterial families followed one of three patterns. Three families maintained a consistent abundance. These included taxa from *Actinobacteria* (*Bifidobacteriaceae* and *Corynebacteriaceae*) and *Firmicutes* (*Carnobacteriaceae*). Ten families decreased in abundance. These included taxa from *Bacteroidetes* (*Flexibacteriaceae*, *Planococcaceae*, and *Prevotellaceae*), *Firmicutes* (*Lachnospiraceae*, *Streptococcaceae*, *Ruminococcaceae*, and *Veillonellaceae*), and *Proteobacteria* (*Bradyrhizobiaceae*, *Moraxellaceae*, and *Sphingomonadaceae*). Five families increased in abundance and included taxa from *Firmicutes* (*Clostridiaceae*, *Enterococcaceae*, *Lactobacillaceae*, and *Staphylococcaceae*) and *Proteobacteria* (*Enterobacteriaceae* and *Xanthomonadaceae*). From a physiological perspective, these results indicate a shift from a microbial community that includes aerobic bacteria that generate energy through respiration to a microbial community that was dominated by anaerobic bacteria that generate energy through fermentation. These bacteria have the ability to form endospores (e.g., *Clostridium*) or ferment lactic acid (e.g., *Staphylococcus* and *Lactobacillus*). Interestingly, most of the taxa that dominated the microbiome throughout decomposition were always at an abundance of at least 2%. This might indicate that endospores from the putrefactive bacteria are present on the skin during the early stages of decomposition, but do not proliferate until the condition of the corpse supports their germination. It also indicates that the lactic acid bacteria on the skin have the ability to proliferate soon after death.

Microbiology, Taphonomy, Decomposition

G76 Swine as a Model for Decomposition: A Comparison of Postmortem Microbiomes

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After attending this presentation, attendees will have a better understanding of the similarities and differences in microbiomes associated with swine and human remains. Swine (*Sus scrofa* L.) are often used as a model for humans in forensic research.

This presentation will impact the forensic science community by determining if vertebrates of different species represent feasible models for human microbial decomposition research.

It is known that microbes are important to the decomposition process and that significant temporal changes in the bacterial community structure occur on carcasses in both terrestrial and aquatic environments.¹⁻³ In this study, human and pig cadavers were sampled daily for five days under identical conditions after placement in the savannah terrain of the Edwards Plateau of Texas (TX-humans and TX-swine). In addition, a comparison to swine decomposition (OH-swine) in the temperate forest of Ohio was considered, where bacteria communities were sampled at 0, 1, 3, and 5 days of decomposition. Sterile cotton swabs were used for sample collection from buccal and skin regions. DNA was obtained from these samples using an organic extraction method. Bacterial variable regions (V1-V3) of the 16S rRNA gene were targeted for amplification using primer pairs 28F and 519R.⁴ The bacterial community structure was determined by modified bacterial tag-encoded FLX amplicon pyrosequencing.⁵ Resulting sequences were processed using Mothur v. 1.29.⁶

There were bacterial genera common to both species, but there were also substantial differences in community structure. In the first study, only 13% of the bacteria were shared between both species and both ecoregions. These primarily included the genera *Corynebacterium*, *Ignatzschineria*, *Staphylococcus*, and *Acinetobacter*. Additionally, 21%, 10%, and 3% of bacteria were shared by TX-swine/TX-human, OH-swine/TX-human, and TX-swine/OH-swine, respectively. Finally, substantial proportions (totaling 54%) of the bacterial species were unique to the species and ecoregion combined; 27%, 17%, and 10% of bacteria were unique to TX-swine, TX-humans, or OH-swine, respectively. The data shows that microbial communities, at the genus level, vary with the ecoregion where decomposition occurs and reveals disparity between bacterial genera from vertebrates of different species. The study demonstrates that each of the two vertebrate species contains its own unique genera and that those populations are influenced by their surrounding environment. More research is needed in this area to determine the potential for swine to be used as prediction models for human and wildlife forensics.

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Sus scrofa, Microbiomes, Decomposition

G77 The Living Dead: An Investigation Into the Internal Microbiome of Human Decomposition

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The goal of this presentation is to demonstrate the unique biodiversity of the bacteria inhabiting corpses at the onset and the end of the bloat stage of decomposition. It will conclude by making comparisons between the internal microbiome and external microbiome of cadavers. It will also demonstrate how abiotic and biotic forces can be influential and can be assessed via remote sensing technologies such as Light Detection and Ranging (LiDAR).

This presentation will impact the forensic science community by progressing the field of forensics by presenting the first data set of bacteria associated with the bloat stage of decomposition. These data represent part of the initial effort to catalog the biodiversity of bacteria associated with decomposition and will add to emerging data sets which will eventually paint a larger picture of the utility of bacteria in forensic research.

Despite the integral role of bacteria in decomposition, few studies have catalogued internal bacterial biodiversity for terrestrial scenarios. To explore this biodiversity, two cadavers were placed at the Southeast Texas Applied Forensic Science (STAFS) facility and allowed to decompose under natural conditions. Each cadaver was sampled at two time points, at the onset and end of the bloat stage, from various body sites including internal locations. This study was conducted at the STAFS facility located at the Center for Biological Field Studies of Sam Houston State University. STAFS 2011-006 was sampled from September 8 to September 15, 2011, and STAFS 2011-016 was sampled from November 3 to November 17, 2011. Cadavers were Caucasian male, not autopsied, and determined to be in early stages of decomposition according to the Megyesi *et al.* Body scoring system.¹ They were placed supine without clothing. Bacteria were collected from 18 locations, three locations during pre-bloat time points (mouth swab,

mouth scrape, and rectal scrape) and six locations during end-bloat points (mouth scrape, small intestine swab, transverse colon swab, sigmoidal colon swab, general body cavity swab, and stomach scrape). Several of the body sites sampled in this study have not previously been sampled from living human beings and are the first of their kind. Bacterial samples were analyzed by pyrosequencing of the 16S rRNA gene following protocols benchmarked as part of the Human Microbiome Project.^{2,3} 16S data was processed and analyzed using QIIME version 1.7.0.⁴ This data also show a shift from aerobic bacteria to anaerobic bacteria in all body sites sampled and demonstrate variation in community structure between bodies, between sample sites within a body, and between initial and end points of the bloat stage within a sample site. The microbiome richness varies widely across cadaver body sites with the most prevalent phyla being the *Firmicutes* and *Proteobacteria*. These data represent the initial cataloging of bacterial species associated with both the pre-bloat and end-bloat stages of human decomposition.

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Bacteria, Cadaver, Bloat

G78 Postmortem Microbial Community Change of Corpse Decomposition on Three Contrasting Soil Types in a Mouse Model System

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After attending this presentation, attendees will understand recent advances in the microbial ecology of corpse decomposition and why it may be important for medicolegal death investigation. Attendees will be presented with results from recent experiments in which microbial community change associated with mouse decomposition on three contrasting soil types was characterized over time.

This presentation will impact the forensic science community by revealing the potential for next-generation sequencing of microbial communities associated with decomposition for estimating the Postmortem Interval (PMI) and locating clandestine

grave sites.

Advances in sequencing technologies have allowed for in-depth characterization of microbial communities across space and time in a wide range of environments. Given that microbes have a nearly ubiquitous distribution and succession-like responses to disturbances, they are excellent targets for tracking the temporal changes in corpse decomposition. However, the feasibility of using microbes for forensic purposes is only just beginning to be rigorously tested. In an initial experiment characterizing microbial community change associated with mouse corpse decomposition on a single soil type, this study concluded that estimating the postmortem interval may be possible by assessing the microbes present at various stages of decomposition. Although these preliminary observations show promise, additional vigorous testing under different conditions is necessary to determine their applicability to forensic science.

This research recently established that bacterial and microbial eukaryotic community data associated with decomposing mouse corpses was sufficiently measurable and directional enough to allow for accurate predictions of PMI as well as the identification of gravesoil. In a follow-up experiment, this study tested how contrasting grave soil-types (e.g., from desert, forest, and grassland environments) affected microbial community change associated with decomposition. It was hypothesized that mouse-associated microbial communities would become progressively different from the starting communities regardless of grave soil type, although late stage corpse-associated microbial communities may be more influenced by underlying soil type. It was also hypothesized that at least some proportion of the soil microbial community associated with late-stage decomposition would be shared despite soil type differences, an important factor for assessing microbes as a forensic tool in grave identification.

A laboratory experiment was performed to characterize microbial communities associated with mouse corpses as they decomposed on three contrasting soil types under controlled conditions for 71 days. Microbial communities (bacterial, archaeal, fungal, and other microbial eukaryotes) were characterized from the skin, abdominal cavity, and gravesoil of five replicate mice at eight time points by sequencing 16S, 18S, and ITS amplicons using the Illumina® HiSeq platform.

This research discovered that microbial communities changed progressively over time at each sample site (skin of head and body, abdominal cavity, and corpse-associated soil). For both abdominal and skin sites, bacterial community change was significantly associated with time, regardless of soil type. Additionally, bacterial communities were initially highly different among soil types, but these communities became progressively more similar as decomposition progressed. For example, *Gammaproteobacteria* in the family *Pseudomonadaceae*, *Betaproteobacteria* of the family *Alcaligenaceae*, and *Firmicutes* of the family *Bacillaceae* increased in abundance in each sample site during decomposition. These trends suggest that a proportion of the soil bacterial decomposer communities are common across contrasting soil types, which makes bacterial communities associated with decomposition very attractive as a forensic tool.

Microbial eukaryotic communities also showed consistent shifts during decomposition with late stage communities dominated by nematodes, amoebae, cercozoa, and fungi. Sequencing of the 18S amplicons revealed that the highest abundance organism in each sample type was nematodes of the Class *Chromadorea*. Samples associated with each soil type were dominated by a different family of *Chromadorea* (desert soil: *Aphelenchidae*; forest soil: *Cephalobidae*; and grassland: *Pangrolaimidae* and *Aphelenchidae*). The amoebae *Heterolobosea tetramitia* was also prevalent in late-stage decomposer communities regardless of soil type, but at highly variable levels. Sequencing of the fungal specific

Internal Transcribed Spacer (ITS) amplicons revealed a shift from fungi in the Phylum *Basidiomycota* and Class *Agaricomycetes* to fungi in the Phylum *Ascomycota* and Class *Sordariomycetes* in late stages of decomposition regardless of soil type. Most of the *Soriomycetes* belonged to the Family *Nectriaceae* and Genus *Haematonectria*.

These results show that postmortem soil microbial communities have great potential as physical evidence because they follow predictable trends.

Postmortem Microbial Ecology, Decomposition, Postmortem Interval

G79 Microbial Diversity in Swine (*Sus Scrofa Domesticus*) Gravesoils Between Seasons

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After attending this presentation, attendees will understand how gravesoil microbial communities may change significantly over a short time period during the summer but not during the winter.

This presentation will impact the forensic science community by showing that postmortem microorganisms during summer investigations should be used differently than postmortem microorganisms during winter investigations.

It has recently been shown that soil microbial communities have potential to serve as physical evidence for estimating postmortem interval. Soil microbial communities respond to the presence of a corpse and the structure of their communities' changes in a predictable manner; however, we are only beginning to understand these changes. It is known that temperature is a significant modulator of corpse decomposition. A corpse typically decomposes more rapidly at warm temperature because insects and microorganisms are more active and chemical reaction rates are more rapid. Yet few decomposition studies have been conducted at colder temperatures when microorganisms and scavengers that drive decomposition and chemical reaction rates are less rapid, if not inhibited. This is a significant gap in knowledge because not all corpses are deposited during the warmer months. To address this gap this study conducted a replicated field experiment to characterize the microbial diversity in gravesoils during summer and winter decomposition.

The experimental unit was a 1.5 x 1.5m plot of pasture located approximately 48km north of Lincoln, Nebraska. The site was enclosed with steel fence to prevent the entry of mammalian scavengers. The climate at the experimental site is a Cold climate type with a mean annual temperature of 9.8°C. Maximum temperatures range from 0°C (January) to 31°C (July). Average annual precipitation is 695mm with approximately 75% of the precipitation between April and September. The soil at the site is a silty clay loam (Mollic Hapludalf) with a texture of 15.1% sand, 53.6% silt, and 31.3% clay. Swine (*Sus scrofa domesticus*) carcasses were placed on February 25, 2008 (winter) and on June 11, 2008 (summer). Control (no carcass) units were also used. Swine carcasses (~20kg) were acquired within 20 minutes of death, which was caused by blunt force trauma to the skull with a bolt gun. Carcasses were placed on the soil surface on their right side facing west within 60min of death. Soil samples (0-5cm depth) were

collected from underneath the carcass at the time of placement, 15 days, 30 days, and 60 days postmortem. The experiment was replicated three times resulting in a total of six carcasses (three summer and three winter).

It was hypothesized that soils associated with pig carcasses decomposing in the summer season exposed to 1,412 Accumulated Degree Days (ADD) would undergo more substantial microbial community change than soils associated with pig carcasses decomposing in the winter exposed to only 325 ADD. Culture-independent, next-generation sequencing approaches were utilized to characterize microbial community change in soil associated with carcass decomposition. Partial 16S and 18S ribosomal RNA genes were sequenced using Illumina® HiSeq and MiSeq sequencing platforms, respectively.

Corpse-associated soil bacterial communities were discovered to have changed significantly compared to control soils during the summer, but not during the winter. The seasonal effect on gravesoil bacterial communities was not simply due to temperature as winter corpse-associated soils with an ADD 326 were highly similar to control soils, while summer samples at an ADD of 322 were marginally significantly different from control soils. Both summer and winter non-carcass control soils were dominated by bacteria of the Phylum *Verrucomicrobia* in the Class *Spartobacteria* and genus *Chthoniobacter*. The late decomposition summer soils increased in abundance of taxa in the Order *Sphingobacteriales* and decreased in the abundance of *Chthoniobacter*.

Microbial eukaryotic sequence data revealed that gravesoil microbial communities associated with summer carcasses became more differentiated from control soils than winter soils that were associated with carcasses. Summer and winter control soils were dominated by similar taxa such as *Cercozoa* in the Orders *Cryomonadida* and *Cercomonas*, fungi in the Classes *Sordariomycetes* and *Agaricomycetes*, as well as the amoeba of the genus *Acanthamoeba*. Late-stage decomposition gravesoils in the summer season showed a dramatic decrease in alpha diversity and became dominated by the cellular slime mold *Fonticula alba* and the endogenous soil bacterivore *Acanthamoeba*. Late stage winter gravesoils had comparable diversity to control soils.

Bacteria, Microbiome, Taphonomy

G80 Quorum Sensing Regulates Blow Fly Attraction and Colonization of Human Remains: Greater Insight Into the Pre-Colonization Interval

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After attending this presentation, attendees will have a better understanding of the mechanisms potentially regulating arthropod attraction and colonization of human remains.

This presentation will impact the forensic science community by illustrating how these data are important as they bridge microbial community ecology and arthropod behavioral ecology as related to the colonization of human remains. Additional research in this area could lead to a great understanding, and potential prediction, of the time interval from death to insect colonization. Furthermore, this research could lead to novel diagnostic tests

allowing for the estimation of the minimum Postmortem Interval (PMI_{min}) by characterizing and quantifying chemicals present on the surface of human remains not colonized by arthropods, also known as the pre-colonization interval.

Arthropod succession is a known process that can be used to estimate PMI_{min} of human remains. However, the mechanisms regulating this process have historically been relegated to arthropod interactions. Data from two research projects will be presented that demonstrate that many of the behaviors exhibited by blow flies (Diptera: *Calliphoridae*) are partially regulated by quorum sensing by bacteria associated with multicellular organisms that colonize and consume human remains, as well as those bacteria intimately associated with the remains themselves. Quorum sensing is the biochemical pathway through which bacteria communicate and make group decisions.

Proteus mirabilis is a common gram-negative bacterium associated with blow flies. This study isolated *P. Mirabilis* from *Lucilia sericata* (Diptera: *Calliphoridae*) and created a knockout library. Strains from this library were unable to demonstrate swarming which is a quorum-sensing response associated with this bacterium. Furthermore, the swarming behavior could be rescued with known fly attractants, indicating a possible relationship between compounds that are known to elicit a quorum-sensing response and fly attraction.

This presentation demonstrates that the lack of a quorum-sensing behavior results in reduced attraction (38%) and oviposition (63%) and that the behavioral responses of blow flies are dependent on their nutritional background, sex, and age. Flies, regardless of age, exhibited a 63% chance of responding to the wild type *P. Mirabilis* than did the mutant. However, older females were more likely to oviposit on the mutant than the wild type, demonstrating a lower level of selectivity. Furthermore, gravid females are ideal models for measuring the impact of bacteria and quorum-sensing compounds on their behavioral responses.

These data are important as they bridge microbial community ecology and arthropod behavioral ecology as related to the colonization of human remains. Additional research in this area could lead to a great understanding, and potential prediction, of the time interval from death to insect colonization. Furthermore, this research could lead to novel diagnostic tests allowing for the estimation of the PMI_{min} by characterizing and quantifying chemicals present on the surface of human remains not colonized by arthropods, also known as the pre-colonization interval.

Quorum Sensing, Blow Fly, *Proteus Mirabilis*

G81 Postmortem Computed Tomography (CT) Observations in Pelvic Fracture: An Unrecognized Pattern of Hemorrhage

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After attending this presentation, attendees will be aware of a previously unrecognized hemorrhage pattern in the retro-sacral area associated with pelvic fracture.

This presentation will impact the forensic science community by documenting a previously unrecognized hemorrhage pattern associated with pelvic fracture that may lead to increased morbidity/mortality.

Methods: Postmortem CT angiography was performed in 25 cases of trauma by selective injections of the iliac arteries. Sites of vascular injury were correlated with pelvic fracture location. A previously unrecognized pattern of posterior retro-sacral hemorrhage was identified and documented at autopsy.

External examination did not indicate contusion/hemorrhage over the sacral area and dissection was initiated based on CT findings. Based on these findings, a retrospective review of 92 consecutive postmortem CT studies was made and analyzed for the presence of a retro-sacral fluid collection. Presence/absence of a retro-sacral fluid collection was correlated with pelvic fracture and other trauma. Cases without evidence of torso trauma served as controls.

Results: In the 25 angiography cases, the pattern of posterior fluid collection and the pattern of pelvic fracture were documented. Contrast extravasation occurred in the distal branches of the internal iliac arteries with tracking into the retro-sacral soft tissue representing hemorrhage. This hemorrhage was related to fractures of the sacrum and separation of sacroiliac joints. In the 92 cases that underwent retrospective review, 14 were eliminated due to soft tissue trauma in the retro-sacral area. Of the 78 cases, 51 had torso trauma and 27 had no trauma. A posterior fluid collection was documented on CT in 29 (62%) of the trauma cases and 3 (11%) without torso trauma. Of the 51 torso trauma cases, 24 (47%) had pelvic fracture and 18 (75%) of these had a retro-sacral fluid collection. In the 27 torso trauma cases without pelvic fracture, retro-sacral fluid collections were present in 11 (41%) of these cases and were associated with fracture of the lumbar spine. In the 3 cases with retro-sacral fluid collection and no evidence of torso trauma, the fluid accumulation was determined to be associated with edema/dependent position.

Conclusion: Using postmortem angiography, retro-sacral hemorrhage in association with pelvic fracture can be depicted. Even without angiography, retro-sacral fluid collection can be identified on postmortem CT. The appearance of a retro-sacral fluid collection was noted in 75% of the cases with pelvic fracture. Unless dissection of the lower lumbar area of the back is performed during autopsy, this collection of fluid would be overlooked. It is important to recognize this finding in order to understand the morbidity/mortality associated with pelvic fracture. Further study is warranted to clarify the specific mechanism for and type of pelvic fracture leading to this retro-sacral hemorrhage.

Postmortem CT, Pelvic Fracture, Hemorrhage

G82New Light in Dead Bodies: Value of Postmortem CT Angiography in Traumatic and Natural Causes of Death

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After attending this presentation, attendees will have gained knowledge about the technical settings, the methodology of postmortem Computed Tomography Angiography (pmCTA), and seen a demonstration of findings in natural and traumatic causes of death.

This presentation will impact the forensic science community by showing the potential of minimally invasive pmCTA as adjunct, or in selected cases, even as a replacement for classic autopsy.

Clinical radiology has led to a great improvement in the diagnosis of vascular pathologies such as traumatic vascular injuries, acute coronary syndrome, pulmonary embolism, or aortic dissection, predominantly based on non-invasive angiographic examinations by means of Computed Tomography Angiography (CTA). Meanwhile, the classical postmortem examination is still

performed according to a method almost unchanged since the time of Virchow, with dissection being the gold standard for determining the cause of death. In order to enhance the customary and partially restricted approach of manual autopsy, the introduction of unenhanced pmCTA in the field of forensic pathology represents a radical innovation, acting as a powerful adjunct or even a potential replacement for classical autopsy. It showed great potential in the depiction of bones and air-containing structures using unenhanced pmCT, but the diagnosis of vascular pathologies remained a "blind spot." At the time of introduction of pmCT at the end of the 1990s, there was no feasible method of applying a contrast medium to the whole adult corpse. During recent years, further attempts to establish a minimally invasive approach using specially designed contrast medium with subsequent pmCTA of the whole body have led to promising results.

This study used an iodinated water-soluble contrast medium mixed with polyethylene glycol (PEG-200) at a mixture ratio of 1:15. This mixture consists of a large polymerized portion (PEG) and an unpolymerized part with small molecular dimensions (contrast medium). After intravascular injection, enhancement is observed for physiologically well-perfused tissues such as the cerebral cortex, myocardium, pancreatic/splenic tissue, renal cortex, liver, and intestinal wall. This enhancement likely arises from the diffusion of the small molecular hydrophilic contrast medium in the capillary bed into the interstitial and intracellular spaces, whereas the polymerized large PEG molecules have to stay in the intravascular space due to their large size. This mechanism on one side avoids the unwanted extravasation of contrast in areas with early decomposition (e.g., the gastrointestinal (GI) tract).

A simple injection protocol is used, consisting of a divided injection of the contrast mixture into the arterial and venous system without establishing new circulation. This technique has proven its potential in more than 200 cases to this time.

As far as could be ascertained, the fastest access to the vascular system for pmCTA is gained by the cannulation of the relatively large femoral vessels. Harvesting of body fluids (blood, urine, Cerebrospinal Fluid (CSF)) for toxicological examination has to be done before angiography to avoid a dilution of the substances targeted in toxicological examination.

There will be an exemplary demonstration of the performance of pmCTA in different body regions:

Brain: Arterial and venous contrast application allows a far better distinction of gray and white matter and the possibility to detect vascular pathologies such as aneurysm or rupture with subsequent hemorrhage than unenhanced pmCT.

Thorax: Reliable assessment of cardiac pathologies and a diagnostic-sufficient filling of its vessels can be shown on pmCTA. Pathologies like aortic dissection are easily displayed by pmCTA. Detecting such a finding prior to autopsy may even lead to a change in anatomic preparation, besides already providing the definitive cause of death.

Abdomen: Despite vascular pathologies, pmCTA allows a very good parenchymal contrast, therefore leading to superior sensitivity and specificity in the detection of organ laceration, rupture, or masses compared to unenhanced pmCT.

Extremities: Even though injuries to the extremities usually do not lead to death, there are several pathologies that need to be detected and may help to reconstruct the inflicted injury, especially in cases of accidents with reconstructive questions or homicide.

The conclusions of this study indicate that pmCTA delivers substantial added value to the classic autopsy regarding vascular and parenchymal pathologies in natural and traumatic causes of death and encourage the adoption of its minimally invasive and relatively inexpensive technique in other institutes of forensic medicine.

G83 Visualization of Myocardial Infarction in Postmortem Multi-Phase CT-Angiography

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After attending this presentation, attendees will understand how the use of Multiphase Postmortem Computed Tomography Angiography (MPMCTA) can help to identify myocardial infarction by detecting a myocardial enhancement after the use of the contrast agent Angiofil®.

This presentation will impact the forensic science community by showing the feasibility of identifying myocardial infarction on MPMCTA. This presentation describes the aspect of the radiological image in comparison with macroscopic and histological findings.

MPMCTA is a minimally invasive technique for postmortem angiography permitting the visualization of the vascular system of the head, thorax, and abdomen in detail. To execute this technique, the cannulation of the femoral vessels of one side is necessary in order to inject the oily contrast agent Angiofil®. After the performance of one native (without contrast agent) Computed Tomography (CT) scan, three angiographic phases are realized: arterial phase; venous phase; and, dynamic phase. For each phase, a CT-scan is executed after the injection of a specific amount of contrast-agent mixture following the standardized protocol of MPMCTA. By comparing the images obtained after the different phases, vascular diagnosis can be performed with very high sensitivity, e.g., in coronary arteries in cases of sudden cardiac death. In some of the cases examined, a pathological enhancement of the myocardium could be observed that seemed to correspond to the morphological finding of myocardial infarction. The goal of this study is to investigate the possibility of identifying a myocardial infarction by MPMCTA.

Twenty-five cases of myocardial infarction (group 1) on which pre-autopsy MPMCTAs had been performed were retrospectively selected. In all cases, the diagnosis was performed by macroscopic and histological analyses. A control group of 25 cases without any macroscopic or histological signs of myocardial infarction (group 2) were then selected. MPMCTA was performed in all cases following a standardized protocol in three phases. The presence or absence of myocardial enhancement and its distribution was investigated by a forensic pathologist together with a board-certified radiologist blinded to the results of previous investigations. This was done first in a subjective manner (enhancement present or not, according to the observers) and second in an objective manner by measuring the mean attenuation in Hounsfield Units (HU) of the myocardium in images of the different phases of angiography.

In all cases of group 1, a pathological enhancement of the myocardium was observed in regions which correlated with the localization of the infarction. Such a pathological enhancement was stated when the mean attenuation was ≥ 95 HU in the arterial phase. In some cases, the enhancement was especially pronounced

(more than 200 HU). This phenomenon was mostly observed in subendocardial regions. In these cases, a chronic infarction could be observed. While the arterial phase mostly showed a diffuse enhancement of the concerned myocardium, it was more concentrated in the subendocardial layer during the venous and dynamic phase. No pathological enhancement was observed in the cases of group 2. The mean attenuation in those cases was ≤ 95 HU in the arterial phase.

By investigating the presence of a pathological enhancement of the myocardium in the arterial phase of MPMCTA with the contrast agent Angiofil®, it is possible to recognize a myocardial infarction. In these cases, mean attenuation in the concerned region is \geq than 95HU in the arterial phase. The following phases can be used to better describe the localization of the infarction and possibly recognize old infarctions.

Postmortem Angiography, MPMCTA, Myocardial Infarction

G84 Investigation of a Disputed Mechanism of Diffuse Axonal Injury Following a Low-Speed Frontal Crash

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The goal of this presentation is to describe an unusual mechanism of a serious brain injury associated with a relatively low-speed frontal crash with an airbag deployment.

This presentation will impact the forensic science community by demonstrating a methodology for investigating an unusual injury mechanism.

Diffuse Axonal Injury (DAI) is a type of widespread traumatic brain injury that is typically associated with higher speed traffic crashes, falls from height, and assaults. The injury consists of shearing of axons in the brain that is associated with a high degree of angular acceleration (rotational forces).

This discussion concerns a single vehicle traffic crash involving a 36-year-old Asian male who was the unrestrained right front passenger of a 1994 Ford® Mustang®. For an unknown reason, the vehicle crossed over the opposite lane, impacted the curb adjoining the roadway, and subsequently struck several small trees on the west side of the sidewalk, coming to a complete stop. The impact speed was estimated to be less than 15mph and there was driver- and passenger-side front airbag deployment. The airbag deployed either during the impact with the curb or upon impact with the thin row of trees just beyond the sidewalk.

The front seat passenger was found unresponsive and was seated in the passenger's seat with his upper torso and head hanging outside the passenger door when police arrived. The passenger was bleeding copiously from a >10 cm laceration over the left posterior aspect of his scalp. The windshield had a classic "spider" fracture on the passenger side, near the top of the windshield where it met the frame, and was bulging outward at the junction of the upper edge of the windshield and windshield frame. The victim was transported to the hospital, where he was diagnosed with a moderate to severe DAI. He was ultimately left with permanent and severe injuries.

An investigation by the insurer resulted in a denial of a claim for the injury based on the following assertion: because the location of the injury to the victim's head was posterior, and the kinematics of a frontal collision would typically involve interaction between the face and top of the head of a front seat passenger and the airbag, and possibly the dashboard and windshield, but *not* the posterior aspect of the head, it was *impossible* for the injury to

have resulted from the crash. The alternative, therefore, was that the injury was caused by an assault that preceded the crash, for which there was no evidence. It was further asserted that, based on experimental primate studies, the speed change of the collision (<10mph) was insufficient to cause the injury.

In the ensuing investigation, it was discovered that there was a significant witness mark inside the vehicle at the top rail above the windshield, indicating a forceful vertically oriented loading of the top rail from below. A review of the CT scan taken on the day of the injury revealed a large laceration that was consistent with a high-velocity tangential load at the left posterior aspect of the head, resulting in a tearing of the scalp forward. Swelling over the right cheek was also documented.

The occupant kinematics and injury mechanism that best explained the evidence were reconstructed as follows: when the vehicle struck the curb, the victim was likely thrown forward and into the dashboard, but the airbag did not deploy until approximately one-half second later, when the vehicle struck the small trees. The deploying airbag then impacted the right side of the victim's face, and propelled his head and torso upward and into the windshield header, resulting in the tangential load that was high enough to deform the metal windshield frame, tear the victim's left posterior scalp, and induce rotational forces sufficient to produce the moderate-severe DAI. Not only was the explanation most consistent with the facts, it was the only plausible explanation for the vehicle damage and witness marks, as no other known mechanism existed to produce the observed damage to the interior of the vehicle. Although an unusual injury mechanism, it was not an impossible or improbable explanation for the evidence, and the assertion that the injury *must* have resulted from an alternative mechanism was rejected.

Diffuse Axonal Injury, Biomechanics, Forensic Epidemiology

G85 Contribution of Antemortem Computed Tomography Findings to Cause of Death Determination: A Case of an Unusual Fatal Stroke

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After attending this presentation, attendees will understand how detailed interpretation of antemortem Computed Tomography (CT) helps to understand the chronology of events leading to death and helps to explore anatomic locations that are difficult to access during autopsy, such as the base of the skull.

This presentation will impact the forensic science community by showing a unique case report in which the antemortem CT was essential to understand the remarkable association of vascular lesions.

Case Report: A 32-year-old man was found fully awake next to his car by a pedestrian, after what appeared to be a car accident. His Glasgow Coma Scale was 15 at the time of initial observation by the paramedics. The initial clinical assessment revealed a facial trauma with a left malar wound and no abnormal neurological signs. He lost consciousness suddenly one hour after initial observation and was intubated. A whole body CT scan was performed and showed a complex left side facial fracture, a metallic

foreign body in the cavernous segment of the internal carotid, associated with an occlusion of the left internal carotid. At first, circulation in the left brain was possible through the Circle of Willis. Twenty-four hours later, another brain CT scan was performed and showed an extension of the occlusion, similar to a carotid artery dissection, ischemic signs in left cerebral hemisphere, a pseudo-aneurysm of the C4 portion of the left internal carotid, and a carotido-cavernous fistula. Twelve hours after this scan, the victim died from a massive stroke of the left cerebral hemisphere.

An autopsy was performed and confirmed that the left malar abrasion was in fact a gunshot entrance wound, the metallic foreign body being a projectile (.22 LR). The left cerebral hemisphere was congestive, edematous, and ischemic. Neuropathology confirmed the anatomical findings of autopsy. Toxicology was negative.

Discussion: The literature provides few case reports of stroke following a gunshot wound. Generally, these occurrences are linked to pellet embolism and the entrance wound is located in the neck area. The vascular lesions concern the common carotid and occasionally the internal carotid. Traumatic carotido-cavernous fistulas are also a rare vascular lesion. Some cases are described in the neuro-surgical literature. The military literature highlights the fact that gunshot wounds associated with internal carotid lesions are very infrequent. According to this research, this case is the first which associates internal carotid dissection, pseudo-aneurysm of internal carotid, and carotido-cavernous fistula. If no antemortem CT had been performed, it would have been difficult to find all these vascular lesions during the autopsy, as this anatomic area (base of skull) is difficult to reach by dissection.

Conclusion: The antemortem brain CT scan permitted better understanding of the chronology of events that lead to death:

- facial gunshot wound
- occlusion and dissection of the left internal carotid due to the projectile in his intra-cavernous portion
- consciousness related to the substitute vascular network (Circle of Willis)
- decrease of intra-cerebral circulation due to extension of the dissection, pseudo-aneurysm, and carotido-cavernous fistula
- ischemic lesions of the whole left cerebral hemisphere
- death due to an increase of the intracranial pressure, edema, and ischemic lesions.

Fatal Stroke, Computed Tomography, Multiple Vascular Lesions

G86 Diffuse Axonal Injury Observed in Structures of the Brain Not Commonly Described

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After attending this presentation, attendees will learn the importance of identifying acute traumatic injuries to the septum pellucidum and fornix when suspected blunt head trauma is entertained. The identification of these injuries in the absence of other fatal head injuries can serve as an important marker for diffuse axonal injury.

This presentation will impact the forensic science community by alerting medical examiners and coroners to other areas in the brain involved by diffuse axonal injury in blunt head trauma cases.

Diffuse Axonal Injury (DAI) is a common traumatic brain injury that causes widespread damage to axonal tracts, as opposed to focal brain injury. It is well documented that DAI results from

acceleration-deceleration and rotational forces. Subsequently, DAI most commonly occurs in Motor Vehicle Accidents (MVA), sports-related injuries, violence, child abuse, and even falls. Diffuse damage to the axons is not caused by the initial mechanical forces but, rather, by secondary biochemical cascades that cause separation of the axons and cell death. DAI is thought to occur over a spectrum of mild to severe injury; a mild form of DAI is commonly referred to as concussion and the most severe end of the spectrum is brain death.

From July 2011 to present, the Santa Clara County Medical Examiner-Coroner Office has collected six documented cases of DAI occurring in areas of the brain not commonly described in the pathologic literature. In two of these cases, subtle findings of trauma were only localized to the fornix and septum pellucidum and in these cases interpreted as DAI. In the other four cases, DAI was recorded in the fornix and septum pellucidum as well as in the typical brain locations previously reported in the literature. The septum pellucidum is a midline structure that runs from the corpus callosum to the fornix and is currently thought to have no functional purpose; the body of the fornix is important for allowing communication between the hippocampus and the hypothalamus. The findings of this study support that these midline structures may serve as a highway of communication from the cerebral hemispheres to vital inner cerebral structures such as the hypothalamus.

All the cases in which DAI was observed are homicides that specifically involve blunt head trauma. The first two cases identified only subtle injuries to the septum pellucidum and fornix in the absence of other dural and brain injuries. Following these two cases, the Santa Clara County Medical Examiner-Coroner Office office has paid more attention to analyzing these inner, midline structures when conducting formal neuropathologic examinations of blunt head trauma victims. In the remaining cases, although sometimes subtle, injuries to these midline cerebral structures were observed where other hallmarks of DAI are also apparent (i.e., cerebral gliding contusions, hemorrhage into the corpus callosum).

In the above cases where death was pronounced at the scene, the microscopic sections did not reveal axonal spheroids as rapid death prevented the necessary interval of hours for microscopic changes to occur. In cases where the survival time was hours, axonal spheroids were visualized even in the absence of β -APP staining.

In summary, the findings of this research illustrate that DAI is being observed in cases of traumatic brain injury in areas not previously or commonly mentioned in the literature. While the literature has yet to describe a function of the septum pellucidum, damage to this structure, as well as the fornix, may be a clear indicator that significant trauma has occurred to the axons in multiple structures of the limbic system and can serve as a marker for diffuse axonal injury when no other fatal injuries are observed. With research showing promise at slowing and minimizing the secondary biochemical cascades that lead to DAI, the ability to clinically diagnose DAI sooner, by focusing on more sensitive locations, can help advance its treatment. The findings of this study will motivate the medical and psychological/psychiatric communities to begin exploring how damage to these specific structures contributes to the debilitating effects of DAI. Such an understanding may have a profound impact on the way living patients suffering from sports-related injuries and veterans returning from the line of duty with head trauma are screened and undergo diagnostic testing and treatment.

Diffuse Axonal Injury, Septum Pellucidum, Fornix

G87 Planned Primary Complex Suicide: Captive-Bolt Gun and Hanging — First Reported French Case

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After attending this presentation, attendees will learn of the first French case of an uncommon form of planned primary complex suicide, the combined application of a captive-bolt gun, and hanging.

This presentation will impact the forensic scientific community by implementing French forensic literature and by illustrating how forensic doctors and crime scene investigators can work together to determine a planned complex suicide.

An exceptional case of suicide with double captive-bolt shots to the head was reported in 2012 by a French team. However, the simultaneous use of a captive-bolt gun and hanging has not been described in France to date.

A complex suicide can be described as one where at least two mechanisms are applied. If the use of these mechanisms is previously intended, then the term "planned" can be used. Moreover, the simultaneous application of the separate mechanisms defines a "primary" complex suicide.

Occupational suicides among farmers in France is unfortunately not an uncommon occurrence. The case of a 58-year-old farmer who committed suicide by captive-bolt gunshot and hanging will be presented. In this case, the victim applied two separate mechanisms to commit suicide. It was apparent that he had carefully planned the simultaneous application of these two mechanisms beforehand, making this case a planned primary complex suicide.

The 58-year-old farmer was discovered in his barn on a late summer evening. His body was hanging from a rope that was tied to a horizontal beam beneath the ceiling of the barn. The body was less than a foot away from a tractor. Shoe and hand prints on the tractor showed that he had climbed on it in order to tie the rope to the beam.

A wound was also found in the occipital region of the farmer's head, and blood flowed from the wound down his back. His feet did not touch the ground, and a metal object was lying less than one meter from them. The object was determined to be a captive-bolt gun. A few blood drops also lay on the ground next to the object. Other blood stains were then found on the edge of a second vehicle, a trailer, which stood more than 2 meters away from the tractor and the corpse. This meant that the farmer had stood on the edge of the trailer and triggered the bolt gun. The measurements carried out on the scene showed that the victim could not have tied the rope to the beam while he was on the edge of the trailer.

This study thus determined that the farmer had first climbed on his tractor to fix the rope to the beam. He then held the rope, climbed on the edge of the trailer, and tightly tied the loose end of the rope around his neck. Next, he held the captive-bolt gun behind his head and shot. The gun instantly pierced a hole in his occipital bone and his body fell off the edge of the trailer and hanged itself. This shows the extent to which the man planned this act. Given that his body could not have remained in equilibrium on the edge of the trailer after the gunshot, hanging occurred immediately, thus defining a primary complex suicide.

Autopsy showed an oval hole in the occipital bone, measuring 1.5cm high and 1.3cm wide. The soft tissue around it showed hemorrhage. Inside the skull was found a subdural hematoma, an intact brain, but with a deep wound of the cerebellum. In the neck region, the right end of the hyoid bone

was found broken. Toxicology reports showed a postmortem level of 2.9g/L of ethanol in blood. With the wound to the cerebellum not being instantly fatal, it was concluded that death was due to a combination of hanging and captive-bolt gunshot to the head.

Complex suicide cases are not common. They are always a puzzle for the forensic doctors who need to carefully analyze the mechanisms before concluding the case was a suicide. Insight can be obtained from precedent cases, and thus the implementation of forensic databases with uncommon cases is useful.

Complex Suicide, Captive-Bolt Gun, France

G88 Utilizing Ultrasonography for Interpretation of Wounds From 40mm Less Lethal Impact Munitions

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The goal of this presentation is to demonstrate the validation of current ballistic testing models to evaluate less lethal impact weapons and how that testing can be applied to predict wound profiling in humans.

This presentation will impact the forensic science community by applying testing models in order to predict wound profiling when the less lethal weapons are applied.

Less lethal impact munitions are law enforcement weapons used to subdue aggressive individuals as an alternative to lethal force. They are also deployed during civil disturbances or other critical events to target specific individuals, such as rioters. The projectiles fired from specialized weapons are intended to deliver blunt trauma similar to a police baton strike, but from a distance. The flight and impact characteristics of some of these projectiles were studied as a result of some negative outcomes from police incidents. It was learned that some manufacturers of these projectiles gather little data as to the effect they may have when striking a human target. When these weapons are deployed in the field, the extent of the wounds they can inflict are generally unknown.

As less lethal projectiles are constructed with a variety of materials, predicting the wound profile they impart can be problematic. Ballistic testing models utilizing calibrated 10% ordnance gelatin and Maki ballistic media have been successfully used to estimate the energy density, impact profile, and potential penetration between a variety of types of less lethal options. The energy transfer into the gelatin can be captured on high-speed video and measured by calculating movement of the gelatin block and velocity loss of the projectile. These measurements are used in conjunction with Maki ballistic media to measure volume displacement of the impact site, similar to "backface signature." Combining the testing data from the media allows comparison between types and models, but it was unclear how the data obtained equates to trauma on living subjects.

During a demonstration of a new 40mm less lethal impact projectile round, police officers volunteered to be exposed to its effects. The lateral mid-thigh was the target area selected for the seven subjects. The subjects also agreed to diagnostic ultrasound scanning of the area before and after impact. Ultrasonography is an imaging modality that has the capability to detect and graph tissue motion and blood flow. Three sets of sonographic images were collected from each test subject and evaluated. When compared to the pre-insult control, the tissue disruption from the less lethal impact was apparent and measurable. The response to the blunt trauma released fluids from the normal spacing within the subcutaneous spaces that were readily detectable. The

scope, depth, and size of the insult was measured and captured. In addition, repeat ultrasound scans were performed at 60 days post event to document the healing process. When the scans were compared to the data retrieved from the testing model, the results were consistent, establishing validity of the previous testing methods to predict wounding inflicted from a less lethal deployment.

Less Lethal, Wounds, Ultrasound

G89 Postmortem Angio-CT in Decomposed Bodies — A Foul Business?

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After attending this presentation, attendees will understand the characteristics in postmortem Computed Tomography-Angiography (pmCTA) in decomposed bodies and know about the general elements in postmortem angiography, as the use of a special contrast agent mixture and the injection technique with a roller pump. Attendees also will learn about the typical minimally invasive inguinal vessel approach and will be familiar with the necessary equipment to perform pmCTA in decomposed bodies. They will also be shown selected case examples.

This presentation will impact the forensic science community by proving that pmCTA is feasible even in heavily decomposed bodies and gains valuable information in addition to an unenhanced pmCT scan. Also demonstrated will be some advantages over conventional autopsy, where preparation and display of pathologies often is hindered by autolytic tissue.

Postmortem cross-sectional imaging has become an important element in forensic investigations over the last decade, primarily by supporting the classical autopsy, which is still considered the gold standard for determining cause and manner of death. Postmortem radiology, especially pmCTA, made great progress in displaying vascular pathologies, such as aortic ruptures or vascular obstructions in fresh bodies, in a minimally invasive manner. However, the detailed diagnosis of these pathologies in decomposed bodies remained a domain of the classical autopsy. Many forensic institutes and medical examiner's offices are privileged to either possess a CT scanner themselves or are able to do postmortem scans on machines in departments of clinical radiology.

A simple injection protocol is used consisting of a divided injection of the contrast mixture, first into the arterial and, in a second step, into the venous system, without establishing "real" circulation. Up until now, this technique has proven its proficiency in more than 200 cases.

According to this research, cannulation of the relatively large femoral vessels is the most reliable and fastest access to the vascular system for pmCTA. Harvesting of body fluids and/or tissue samples (blood, muscle, urine, Cerebrospinal Fluid (CSF)) for quantification of substances in the toxicological examination has to be done before angiography to avoid a dilution of the substances targeted. Harvesting of kidney samples in the absence of urine can be done during autopsy, since the dialysate of kidney samples is used for toxicological pretests only (solely qualification of substances, no quantification).

Other forensic pathologists and radiologists are encouraged to become acquainted with this simple, comparatively low-cost and yet significant method in forensic imaging.

G90 Possibility of Postmortem Interval Determination From the Analysis of Degraded DNA of Cadaver Tissues Via Flow Cytometry

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After attending this presentation, attendees will be able to understand the biochemical processes that DNA undergoes after death. Analysis by flow cytometry will show if this process is constant over time and what role temperature and cause of death have in arriving at conclusions.

This presentation will impact the forensic science community by demonstrating the role of flow cytometry and its use to aid in death investigations by providing a more accurate and reliable method with which to calculate the postmortem interval when the death event is recent.

Determining the time-since-death is one of the most important questions that need answering when conducting death investigations. Through the years, many methods have been formulated to deduce this data, but no method is deemed absolute. Temperature-based methods, involving the use of liver and rectal thermometers as well as Henssge's nomogram, appear to be the most widely used. Other methods, such as liver mortis, rigor mortis, and decomposition stage, not only require visual observations to be taken into account, but the physical state of the corpse must also be noted. When using these methods, other external factors must be taken into account, which further complicates the calculation of the postmortem interval. The problem arises when human observations of the physical data must be utilized to arrive at a reasonable conclusion. Human error opens the doorway to questionable doubt as to the accuracy of the postmortem interval determination. Forensic entomology can provide definitive results, but its use is still dependent upon the ability of the insects to gain access to the corpse in a timely fashion. While analysis of the vitreous humor takes note of biochemical changes which occur within the eyeball, the use of this method is limited to only a few causes of death.

In this study, determination of the reliability of calculating the postmortem interval based upon the analysis of necrotic brain and cardiac tissues using Flow Cytometry (FCM) to map a cellular pattern which correlates directly to the death event was researched. Use of the FCM allows for the almost instantaneous analysis of thousands of cells, resulting in data which is unbiased, making it ideally adaptable for the study of cell death. This study will use cadaver tissues collected from 20 random autopsy cases performed by the medical examiner at Alabama Department of Forensic Sciences Montgomery Medical Laboratory. Information about the age, race, sex, and cause of death will be recorded, as well as the known time of death. The study will be conducted in two temperate conditions (4°C and 21°C) to ascertain the role of temperature in the cellular degradation process and its effects on the resulting data conclusions. Previous studies have found the use of flow cytometry to study other organ tissues to be viable up to 72 hours; however, this study hypothesizes that the use of brain and cardiac tissue is more durable and reliable, depending on the cause of death.

Postmortem Interval, Cadaver Tissues, Flow Cytometry

G91A Multidisciplinary Forensic Effort Unwraps the Mystery of a Mummified Case

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After attending this presentation, attendees will understand the effectiveness of a multidisciplinary approach in solving difficult forensic cases.

This presentation will impact the forensic science community by presenting the options and resources available to medical examiners for dealing with severely mummified remains. In this instance, experts from multiple fields may be necessary to arrive at a conclusive opinion as to the cause and manner of death.

On October 12, 2012, the mummified remains of a 23-year-old Black male were discovered in the mobile home where he lived with his mother and 18-year-old stepsister. A pungent odor had been reported coming from the mobile home. The decedent was severely mentally challenged, suffering from Cri du Chat (chromosome 5 deficiency). His mother, the primary caregiver, reported that he died in February 2012. The remains were transported to the Georgia Bureau of Investigation's Eastern Regional Crime Lab for examination.

The severely mummified body was clad in a soiled disposable adult diaper, weighed 12 pounds, and was covered with innumerable dead larvae and pupa cases. The medical examiner found no evidence of attempted resuscitation, medical therapy, or recent injury; however, the spine had steel rods that were used to correct neuromuscular scoliosis. An internal examination was not possible because of the advanced state of mummification and the fixation of the body into fetal position; therefore, the medical examiner contacted a forensic anthropologist to examine the remains. The fragile tissues were rehydrated using a room temperature water bath and then carefully dissected. The anthropologist X-rayed and weighed the remains post-processing to assess bone density. Skeletal epiphyses were examined to determine skeletal age, and the long bones were measured to compare the decedent's bony dimensions to those of healthy Black males of similar age. Skeletons from the University of Tennessee's Bass Donated Collection were used for bone weight comparisons, and data from the Forensic Data Bank was used for osteometric comparisons. Samples were also taken from the bed linens upon which the body was discovered in order to approximate Postmortem Interval (PMI) using the Vass *et al.* (1992) Volatile Fatty Acid (VFA) method, as no literature was available to support a PMI estimate of >1 month given the unique conditions in which the body decomposed. These samples were submitted to a biochemist for VFA extraction and analysis.

The skeletal remains exhibited a number of congenital and pathological anomalies. Pathological anomalies included delayed skeletal maturation, craniosynostosis, severe dental decay, advanced osteoporosis, porotic hyperostosis, and cribra orbitalia. The skeleton weighed 4 pounds post-processing (including the Galveston rods), compared to an average skeletal weight of 10 pounds for healthy Black males aged 18-30 years. Skeletal age based on epiphyseal union was determined to be 18-23 years. The state of most epiphyses (specifically shoulder and pelvic girdles) pointed to an age at the low end of this range. The decedent's bone dimensions were consistently within the lowest percentile

(1%) or completely outside of the range of normal variation for adult Black males. These measurements were input into the FORDISC® 3 program to evaluate typicality, and the program classified the decedent as a Black female (on account of the small size). The VFA results were inconclusive, as the time lapse between death, collection, and analysis was too long to get an accurate profile of the volatiles. Alternative analyses of the bedding samples are ongoing.

A review of the medical literature suggests that Cri du Chat syndrome symptoms include feeding problems because of difficulty swallowing, low birth weight, poor growth, scoliosis, and Gastroesophageal Reflux Disease (GERD). The decedent's medical records revealed that he exhibited a number of these symptoms. He had undergone fundoplication for his GERD and was scheduled to have a redo of this surgery and insertion of a gastrostomy tube in 2004 (eight years prior to death); these were the last medical records available for review. While delayed maturation can be attributed to Cri du Chat, and osteoporosis and low bone density in the load-bearing skeletal elements can be attributed in part to immobility, porotic hyperostosis and cribra orbitalia are associated with significant nutritional deficiencies (i.e., severe anemia) and are rarely encountered in modern populations. Based on the medical records, investigative reports, and anthropology investigation, the medical examiner ruled the cause of death as complications of medical neglect and the manner of death as homicide.

Mummified Remains, Forensic Anthropology, Cri du Chat

G92 Skeletal Pathology With Associated Soft Tissue Defects Inform Physiognomy

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After attending this presentation, attendees will understand how skeletal pathology can be used to infer associated soft tissue pathology to better provide a physical description of a decedent to aid in identification of nearly skeletonized remains. Presented are the characteristics of Klippel-Feil Syndrome and a case study of identification aided by a profile of the common presentation of this syndrome.

This presentation will impact the forensic science community by serving as an example of interdisciplinary collaboration for developing a biological profile and identification.

When conducting a postmortem evaluation, pathologists note skeletal abnormalities that are readily visible on radiograph or noted in dissection; however, their significance may be overlooked unless related to cause of death. However, when examining unidentified skeletal remains, these abnormalities may provide clues related to the soft tissue physiognomy of the decedent and/or associated defects. These findings can then be combined with other aspects of the biological profile to provide a better description of the decedent, including possible conditions that would be apparent in the decedent's appearance that law enforcement can then use to search for possible putative decedents.

This study presents a case of Klippel-Feil syndrome, a rare skeletal abnormality (1 in 42,000), marked by congenital failure of vertebral segmentation, resulting in fusion of at least two cervical vertebrae with other skeletal changes often noted. The most typical presentation of this syndrome is fusion of C2 and C3, but fusions of any sub-set of cervical vertebrae are possible and, in some instances, fusion of thoracic or lumbar vertebrae have been noted. Additional skeletal changes include the increased incidence of scoliosis, transitional vertebra, rib abnormalities,

cleft palate, cranial asymmetry, and short stature. Aside from the skeletal presentation of this syndrome, there are numerous soft-tissue or physical presentations that may have been noticeable to family and acquaintances of the decedent. Signs of Klippel-Feil most often include a low posterior hair line, a shortened neck, and limited range of motion of the neck. Additional conditions that may be noted include hearing problems, kidney and heart conditions, shortened stature, webbed neck and digits, and elevated scapulae. The common presentations of this syndrome were used to develop a profile that was utilized by law enforcement in their investigation to establish a presumptive identity and to secure medical records for a putative decedent. Once medical records are located, personal identification is accomplished via antemortem/postmortem radiographic comparison. In these cases, the radiographic comparison is greatly aided by the unusual morphology secondary to the pathological condition.

Skeletal Pathology, Klippel-Feil Syndrome, Forensic Identification

G93 Structured Light Illumination (SLI) Capture of Postmortem 3D Fingerprint Point Coordinates

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The goal of this presentation is to introduce the audience to Structured Light Illumination (SLI) Capture and its potential application to the process of postmortem identification of decomposed remains.

This presentation will impact the forensic science community by introducing the potential application of SLI Capture to the process of postmortem identification of decomposed human remains, thereby effecting a more rapid identification of unidentified bodies, potentially increasing the solve rate for homicides.

The identification of unidentified human remains in the nation's medical examiners' and coroners' offices has been called a silent epidemic. Many of these cases remain open due to a failure to associate a putative identification between a known set of remains and a missing person. Technology such as the National Missing and Unidentified System (NamUS) protocol have been utilized toward the end of increasing the number of leads between bodies and specific missing subjects. Once associations have been made, screens allow specific identification between subjects to variable degrees of certainty, based on the methodology employed. All methods of identification have limitations: dental is scientific but relies on proper records; DNA is highly reliable but time-consuming and expensive; fingerprints are reliable but require known prints; and so forth. All methods are adversely impacted to varying degrees by decomposition. One of the earliest postmortem changes with decomposition is loss of useful fingerprints on a set of remains. This may occur with loss of printable surface as a result of slippage of the epidermal glove, distortion by prolonged water submersion, desiccation through mummification, etc. In many instances, visible fingerprint information is present but not forensically useful for identification purposes due to the inability to accurately collect, utilize, and maintain such data. Many of these unidentified remains are homicide victims and the lack of or delay in victim identification adversely impacts law enforcement investigative efforts, lowering the total number of cases solved.

In an effort to find a possible solution to the issue of

obtaining fingerprints in cases where conventional efforts are difficult, impractical, or impossible, a prototype digital screening process for the examination and comparison of low-quality postmortem fingerprints was created. The system employed utilized a SLI technology to capture the full three-dimensional point coordinates of the fingerprints with a native resolution of ~1,000 pixels per inch (ppi) with a final flattening resolution and uniform spacing of 500 ppi. SLI uses a series of projected light patterns captured at an angle by a camera. The distortion of the patterns off the target surface is used to calculate the surface point coordinates in three-dimensional. SLI methodology is well-established in research and industrial applications. Within biometrics and forensics, SLI has allowed direct measurement of the fingerprint ridges on living subjects. For postmortem fingerprint capture, the SLI technique captures the surface topology and ridge patterns via penetration of the light patterns and interference with structures within the tissue

The prototype instrument was deployed into an operational medical examiner's office, with cases selected on a prospective basis. Inclusion criteria were based on gross examination of the remains' fingers by a single pathologist in determining whether or not the system might be useful to enhance otherwise suboptimal fingerprints. Optical scans were conducted and digitally stored for subsequent single-blinded comparison by a certified latent fingerprint examiner.

Preliminary results document that useful postmortem fingerprints can successfully be utilized for identification purposes in cases where conventional techniques may not be as effective. Benefits of the system include the potential for increased identifications based on the inexpensive, easy, rapid, and reliable methodology of postmortem fingerprints. An additional benefit is that fingerprinting can be conducted without physically contacting the decedent's body, thus preserving potential trace evidence while allowing scientific confirmation of identification prior to autopsy examination. Miniaturization of the prototype will allow such processes to be conducted on scene, before a body is transported. The hope is that, ultimately, increasing rapidity of identification will expedite case investigation in homicides and improve solve rate.

Structured Light Illumination, Decomposition, Fingerprints

G94 Fatal Hemorrhage From Varicose Veins Resembling an Accidental/Traumatic Cause of Death

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After attending this presentation, attendees will be able to recognize common findings in cases of fatal hemorrhage from lower extremity varicose veins, including demographic information and potentially misleading blood patterns at the death scene.

This presentation will impact the forensic science community by showing the importance of medicolegal death investigators and forensic pathologists recognizing that hemorrhage from lower extremity varicose veins — a common and usually benign disease process — can be fatal and should be distinguished from an accidental/traumatic or even homicidal cause of death.

Varicose veins are a common and generally benign disease process usually affecting women more than men. Varicosities are caused by defective venous valves, resulting in increased venous pressure and dilation of the veins; often the subcutaneous veins of the lower extremities are affected due to an increase in blood

pressure in the standing position. Symptoms can range from a cosmetic problem to pain and deep venous thrombosis, and in severe cases may lead to chronic ulceration and hemorrhage.

This report describes the case of a 78-year-old man who had been in town visiting family for the past ten days. The decedent had a history of hypertensive cardiovascular disease and atrial fibrillation with remote pacemaker placement and recent (approximately 2 weeks prior) pacemaker replacement. He was functional and active, spending much of his time traveling across the country. The decedent was last known alive around 11:45 p.m. the prior evening following a late dinner. About one hour later, his daughter and son-in-law awoke to strange sounds coming from inside the home and upon investigation found the decedent unresponsive at the top of the stairs of the basement in which he had been staying. A large amount of blood was at the death scene with pools of blood on the floor at the bedside, near a phone that was disconnected from its receiver, and near a sofa. A trail of bloody right footprints seemed to decrease in intensity as the trail appeared to progress from the bedroom to an adjoining room and up the stairs.

Autopsy findings were negative for traumatic injuries of the body. There was stasis dermatitis of the lower legs with an abundance of cutaneous varices and a few circular areas of scab formation surrounded by various stages of fibrosis. Further inquiry into the decedent's medical history revealed he had a history of bleeding from his foot or ankle, most recently about a week before his trip. A few years prior, he had a bleeding episode which began in the shower and was severe enough to require assistance from his daughter-in-law. Another bleeding episode of the foot occurred approximately two months prior to this trip, after which the decedent sought medical attention from his primary care physician. At that time, his primary care physician recommended a vascular surgery consultation since the bleeding episodes were becoming a recurrent problem. It is not known whether the decedent had followed up on this recommendation. The cause of death was certified as exsanguination due to bleeding cutaneous varices of the right ankle. Other significant conditions included hypertensive and atherosclerotic cardiovascular disease with atrial fibrillation status post-pacemaker placement. The manner of death was ruled as natural.

Fatal hemorrhage from lower extremity varicose veins is a rare occurrence. Cases reported in the literature share some features: the decedent is oftentimes elderly with a history of cardiovascular disease and is discovered deceased after an apparently unwitnessed event. A large amount of blood is present at the scene due to increased pressure from a venous system, which may resemble arterial bleeding. Investigative and autopsy findings are negative for trauma and positive for venous stasis changes and cutaneous varices of the lower extremities.

Varicose Veins, Blood Pattern Analysis, Investigation

G95 Methadone in Methadone-Related Deaths and in Impaired Drivers: Comparative Study of Results of Toxicological Analyses

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After attending this presentation, attendees will learn about postmortem methadone interpretation, that concomitant drug intake is very frequent in methadone-related deaths, and that the drugs most frequently involved are prescription drugs such as benzodiazepine and neuroleptics, which may prolong the QT interval. Drugs acting on central opioid receptors and cocaine are less frequently observed in postmortem cases than in impaired drivers.

This presentation will impact the forensic science community by increasing understanding and interpretation of methadone-related fatalities and showing that there is a need to perform more studies in order to further understanding and prevent methadone fatalities, especially during substitutive programs. Complete toxicological analyses are essential to understand the role of concomitant prescription drugs intoxication.

The goal of this study was to evaluate methadone-related deaths and to determine whether differences exist between the concomitant drugs found during toxicological analyses between methadone-related deaths and impaired drivers under the influence of methadone.

Materials and Methods: Methadone-related cases were reviewed retrospectively on autopsy reports concerning methadone-related deaths performed at the University Centre of Legal Medicine in Lausanne from 2000 to 2010 and on results of toxicological analyses of impaired drivers under the influence of methadone controlled between 2000 and 2007. For postmortem cases, complete autopsies, including histological examination and full toxicological screenings, were performed. The urine was screened for illicit drugs using immunoassays. Comprehensive drug screenings were performed on peripheral blood and urine by Gas Chromatography/Mass Spectrometry (GC/MS). Drug screenings were also performed on peripheral blood by High-Performance Liquid Chromatography coupled with Diode-Array Detection (HPLC-DAD) and by Headspace-Gas Chromatography-Flame Ionization Detection (HS/GC/DIF) for the detection of volatile substances. The confirmation and quantification of drugs was done by GC/MS, GC/MS/MS, or HPLC/MS/MS. Then a comparative study of toxicological analyses between both groups was performed.

Results: A total of 126 postmortem cases were selected (89 men and 37 women). The age of the victims ranged between 15 and 57 years with the mean age of 33 years. In the group of drivers, there were 148 cases (133 men and 15 women) with the age ranging between 19 and 58 years and the mean age of 34 years.

The median methadone blood level was 395µg/L in the postmortem group and 147µg/L in the drivers' group. Methadone was detected in blood without any other substances in one postmortem case and in five driver cases. Benzodiazepines were the most frequently observed concomitant drugs (78% in the postmortem group and 53% in the drivers' group). The distribution of other concomitant drugs was as follows: drugs acting on central opioid receptors (33% in the postmortem group and 57% in the drivers' group), QT-acting drugs (37% in the postmortem group, and 12% in the drivers' group), cocaine (33% in the postmortem group and 52% in the drivers' group), and ethanol (21% in the postmortem group, and 23% in the drivers' group).

Discussion and Conclusion: The determination of whether or not a death is related to methadone is controversial because therapeutic and toxic blood levels overlap and others drugs are frequently found during toxicological postmortem analysis. This study's results are in accordance with current clinical guidelines suggesting a higher risk of sudden death when methadone is administered with other drugs. This study illustrates that methadone-related deaths could be due to the toxic effects

of other drugs acting on the central nervous system (respiratory depression) or due to cardiac arrhythmias. More postmortem studies should be performed in order to further understand and prevent methadone fatalities, especially during substitutive programs.

Methadone, Benzodiazepines, Long QT Drugs

G96 Firearm-Related Deaths in Tarrant County, Texas

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After attending this presentation, attendees will have learned the incidence of gun-related deaths and the proportion related to manner, age group, gender, and race/ethnicity in Tarrant County, Texas, from January 2006 to December 2012 and its comparison to larger studies.

This presentation will impact the forensic science community by increasing understanding of the types of gun-related deaths and the demographics most commonly affected by gun violence.

Introduction: Over 30,000 people die annually in the U.S. from injuries caused by firearms.^{1,2} Texas ranked 23rd among states for firearm deaths in 2010 with a rate of 11 deaths per 100,000 population, which is higher than the U.S. average of 10.1 deaths per 100,000 population.¹ Tarrant County had a rate of 10.8 firearm deaths per 100,000 population, which is slightly lower than Texas but higher than the national average. The issue of gun control is a current popular topic given the recent national tragedies. Regardless of personal or political opinion, firearm deaths affect every demographic and it is important to recognize the distribution in order to identify strategies to reduce them.

Material and Methods: This study included all firearm-related deaths in Tarrant County, Texas, from January 2006 to December 2012. The data was collected from the Tarrant County Medical Examiner's Office records retrospectively. The deaths were grouped by manner and were analyzed according to age group, gender, and race/ethnicity, then compared to national data obtained from the Centers for Disease Control and Prevention's (CDC's) web-based Injury Statistics Query and Reporting System.¹

Results: There were a total of 44,495 deaths recorded in Tarrant County, Texas, from January 2006 to December 2012 and 1,161 of these deaths were related to the use of firearms (2.6%). Nationally, only 1.3% of deaths were related to guns between 2006 and 2010. The largest portion of gun deaths were suicides (60% of gun deaths, 53% of all suicides), followed by homicides (37% of gun deaths, 53% of all homicides), accidents (1% of gun deaths, 0.4% of all accidents), and undetermined (1% of gun deaths, 5% of all undetermined deaths). Males were more likely to die by firearms compared to females (82% males, 18% females). There was a higher incidence of gun suicide than homicide in males and the reverse was true of females. The majority of gun deaths occurred in those 19-65 years of age (83%) and manner was usually suicide or homicide. The deaths that occurred between those 0-18 years of age were primarily homicides while those greater than 65 years were largely suicides. Compared to the overall U.S. population, Tarrant County has a smaller Caucasian and larger Hispanic population; however, Caucasians accounted for 63% of gun deaths followed by African Americans (19%), Hispanics (14%), Asians (4%), and Native Americans (0.2%). Caucasians accounted for the majority of suicides by gun (85%); however, homicide victims were

mostly African Americans (40%) followed by Caucasians (28%) and Hispanics (25%).

Conclusion: Tarrant County has a larger incidence of suicide by gun (53.1%) compared to the U.S. (50.5%) and less homicide by gun (53.8%) compared to the U.S. (68.5%). In both, suicides are the most common manner of death in relation to guns, accounting for 58% of gun deaths and more than 50% of all types of suicide. There have been major achievements in other safety issues such as tobacco cessation and motor vehicle accident prevention and it has been proposed that similar tactics could be applied to firearms, such as: (1) a public-awareness campaign to increase gun safety and recognize at-risk individuals; (2) limit the depiction of gun violence on TV and video games; and, (3) take safety measures to limit access to appropriate users, etc.⁵ There has been a lack of rigorous research on firearm fatalities since 1996 due to a bill restricting CDC funds on research that would advocate or promote gun control.⁶ However, recently the CDC was granted funds to conduct further research on the causes and prevention of gun violence.⁶ Research can have lasting effects as shown by the decrease in motor vehicle, fire, and drowning fatalities over the last 20 years, which was accomplished without banning these things but instead from translating findings into effective interventions.⁴ Identifying the at-risk demographics will help to create better solutions.

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Firearms, Guns, Death

G97 Sudden Death in a 6-Year-Old Due to a RyR2 Mutation Associated With Catecholaminergic Polymorphic Ventricular Tachycardia (CPVT)

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After attending this presentation, attendees will have an understanding of the clinical features and pathophysiology of a RyR2 mutation associated with Catecholaminergic Polymorphic Ventricular Tachycardia (CPVT).

This presentation will impact the forensic science community by raising awareness that sudden unexplained death may be caused by an undiagnosed cardiac channelopathy and that funding for genetic testing is needed in medical examiner jurisdictions with budgetary constraints.

A 6-year-old boy with no antecedent complaints of illness was found in cardiopulmonary arrest on the bathroom floor at school. Subsequent advanced resuscitative measures failed. Further investigation revealed no family history of sudden unexplained deaths or known cardiac abnormalities. The decedent had a negative past medical history with no history of dizziness, syncope episodes, or palpitations, and by all accounts had been a normal, active 6-year-old boy. At autopsy, external examination was unremarkable with no evidence of injury. Internal examination revealed no anatomic cause of death. The heart was anatomically normal with biventricular dilation. Histologically, thin, stretched myocytes with groups of wavy fibers consistent with the dilation seen grossly were present. The conduction system was unremarkable. Acute contraction band necrosis of immediately subendocardial cardiomyocytes as well as deep within the papillary muscles was present microscopically. Complete histological examination of all organs as well as toxicological, biochemical, and microbiological studies revealed no cause of death. No funding to obtain genetic testing for cardiac channelopathies was available; however, the parents expressed a willingness to procure financing for these studies with hopes of determining a definitive cause of death. Genetic testing for cardiac channelopathies was obtained and a RyR2 Ser2246Leu Class I mutation was detected which is associated with CPVT. The RyR2 gene encodes the ryanodine receptor 2 protein which forms channels controlling release of calcium ions from the sarcoplasmic reticulum into the cytoplasm which activates cardiomyocyte contraction. Over 70 mutations of the RyR2 gene causing CPVT have been found. Individuals with CPVT may develop polymorphic ventricular tachycardia during exercise or intense emotion which may self-correct or degenerate into ventricular fibrillation with subsequent death. Age of onset is typically from 7 to 9 years, but initial manifestation of CPVT has been known to occur in adults as well. RyR2-related CPVT is inherited in an autosomal-dominant manner which prompted genetic testing of a sibling and the parents. This presentation discusses the clinical features and pathophysiology of a RyR2 mutation associated with CPVT, indications for genetic testing in sudden unexplained death, and the fiscal challenges in obtaining such testing.

RyR2 Mutation, Channelopathy, Sudden Death

G98 Multiple (Eight) Self-Inflicted Gunshot Wounds to the Head

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After attending this presentation, attendees will understand that autopsies involving multiple gunshot wounds to the head, even though routinely approached as a homicide based on the circumstances, scene investigation, autopsy, and crime lab testing, could be self-inflicted.

This presentation will impact the forensic science

community by generating awareness toward working up a case of multiple gunshot wounds based on the circumstances, history, and scene investigation.

Cases of multiple (considered to be more than two) self-inflicted gunshot wounds are rare and require careful examination of the scene of occurrence, thorough consideration of the decedent's psychiatric, medical, and social histories, and accurate postmortem documentation of the gunshot wounds and their trajectories through the body.

Lethal self-inflicted gunshot wounds are most commonly sustained to the head (81%), often in the right and/or left temple, followed by the mouth and submental region of the neck, the chest (17%), particularly the precordial region, and the abdomen (2%).^{1,2}

In this case, the decedent had a history of depression, without evidence of prior suicidal ideation. At the scene, a six-shot .22 caliber ROHM® RG7 handgun was recovered under the decedent's body and the decedent's blood was present on a lock box in the residence that contained firearm ammunition. Postmortem examination of the body revealed eight entrance gunshot wounds on the decedent's head. There was no evidence of close-range firing (i.e., soot deposition or gunpowder stippling) surrounding any of the entrance defects. Two of the gunshots penetrated the skull, causing injury to the brain. The remaining gunshots were associated with fragmented bullets recovered in the scalp adjacent to the skull. Fingerprint analysis, blood stain analysis, and test firing patterns were evaluated on the gun recovered at the scene.

Upon review of the literature pertaining to multiple self-inflicted gunshot wounds to the head, most cases document two gunshot entrance wound defects. In a case reported in 1906, a subject shot himself seven times and survived, later committing suicide by hanging. The current case portrays the greatest number of self-inflicted gunshot wounds to the head (eight) resulting in death, compared to the published literature.

Lethal self-inflicted gunshot wounds are most commonly sustained to the head (81%), often in the right and/or left temple, followed by the mouth and submental region of the neck, the chest (17%), particularly the precordial region, and the abdomen (2%).

It is a common misconception that multiple gunshot wounds of the head are invariably homicidal. In some instances, multiple gunshot wounds of the head are not lethal. Karger explains that an individual's capacity to act following one or more gunshot wounds of the head may be due to the preservation of areas in the central nervous system referred to as "targets of immediate incapacitation." Such targets include the upper cervical spinal cord, brainstem, cerebellum, diencephalon and midbrain including the basal ganglia, the motor cortex, and the capsula interna.³ Following multiple gunshots to the head, an individual's faculties may be fully preserved or minimally limited, if the central nervous system is not injured by the gunshot wound, either because the bullet did not penetrate the skull or because the bullet entered the cranial vault without injuring vital targets.⁴⁻⁶ In the case at hand, only two of eight small-caliber projectiles entered the cranial vault. The victim was clearly not incapacitated early in the course of events as he was able to open the lockbox, while bleeding, and reload the weapon.

Autopsies involving multiple gunshot wounds to the head should be approached as homicides until proven otherwise. However, depending on the scene findings, clinical history, and lethality of injuries, suicide should not be excluded out of hand.

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Self-Inflicted, Multiple, Gunshot Wounds

G99 Cognitive Bias in Forensic Entomology: A Life History Approach to Assessing Impact

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After attending this presentation, attendees will have a better understanding of how the most common types of cognitive biases influence decisions made throughout multiple stages of a forensic entomological case life history.

This presentation will impact the forensic science community by bringing awareness of cognitive biases and when they are likely to influence decision making.

Of central focus is the proposition that expectations formed before and during problem solving place all forensic scientists in especially vulnerable situations for biased conclusions of which they are not consciously aware. This presentation maintains that an awareness of these biases, and when they are likely to influence decision making, is paramount to elevating forensic science in the direction of more sound scientific methodology. In this presentation, attention is focused upon forensic entomology as an example of a discipline not previously connected with cognitive bias in the literature, but just as open to problems in this area as any other discipline.

Much of what forensic scientists do throughout a case life history falls under the cognitive science areas concerned with perception and problem solving. Whether the procedure involves fingerprint analyses, anthropology, or voice recognition, forensic scientists are often called upon to make decisions based on pattern recognition (more specifically, signal detection). It is widely held in the cognitive literature that in most situations involving perception, the brain adopts a top-down mode of processing, one where the interpretation is "constructed" from memory and highly influenced by context. This default mode of the brain puts all forensic scientists at risk of forming biases throughout perception and problem solving, even under normal circumstances. When the perceptual task is applied to a forensic context, the consequences of mistakes in perception are far more dire, and as will be argued, far more likely to go unnoticed.

Cognitive biases are the result of expectations formed before and during problem solving. They are almost always made without conscious intent or awareness. This study maintains that the four types of cognitive biases most pertinent to forensic procedures involve: confirmation bias; evaluation bias; selective reading; and semantic priming. In confirmation bias, all evidence which refutes our initial hypothesis is systematically minimized or ignored. In evaluation bias, decisions are influenced by what is

considered the most desirable outcome to occur at the conclusion of the analysis. In selective reading, the "gist" of a written passage conforms to preconceived interpretations and not what is actually presented. In semantic priming, previous contexts or experiences prime the brain to perceive ambiguous stimuli in specific and predictable ways.

When applied to a case life history, this presentation will demonstrate how and when each of these four cognitive biases influence perception and decision making for the forensic entomologist from the point of first becoming aware of the case through witness preparation and testimony. Further, modifications to the scientific method will be presented that could be adopted to overcome some of these cognitive biases. The conclusion is that while forensic entomologists are most vulnerable to confirmation and evaluation biases throughout a case life history, selective reading and semantic priming can influence decision making at crucial stages. While the specifics of the case life history model adopted here focus on the forensic entomologist, they can easily be expanded to most, if not all, forensic scientist case life-histories.

The more knowledgeable the forensic community is regarding the influence of cognitive biases, the better prepared programs will be in training forensic students. In its current state, members of the forensic science community gain a reputation based on the number of cases where the evidence they collected, analyzed, and summarized for the court leads to a conviction. Nowhere in the process is science able to evaluate whether the conclusions drawn from the evidence were correct. A focus on training methods which teach individual scientists when to be aware of potential cognitive bias is vastly more ideal. The long-term ramifications of a community of forensic scientists who have been trained to be skeptical of their decisions can only strengthen the field and lead to better science presented in court and better verdicts made by juries.

Cognitive Bias, Perception, Diptera

G100 Drownings in a Desert State: Who? Why? How?

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After attending this presentation, attendees will understand the demographics, circumstances, risk factors, and common autopsy findings in deaths due to drowning in a desert state (New Mexico).

This presentation will impact the forensic science community by highlighting the risk factors for drowning deaths and suggesting possible public health avenues for mitigation of these deaths.

Despite having less than 1% surface area of water and fewer than 100 public swimming pools, New Mexico consistently ranks as having one of the highest per capita rates of drowning deaths. Better understanding of the populations most at risk and appreciation of the most common autopsy findings not only may improve diagnosis of a notoriously difficult cause of death (drowning) but also may help to develop public health strategies specifically geared toward preventing drownings in desert states.

A retrospective review of drownings via query of the statewide New Mexico Office of the Medical Investigator electronic database yielded over 400 deaths during 2000-2010. Across all ages and races, males were vastly over-represented in drowning deaths (74%). This is similar to that reported nationally.¹ Median age of both male and female decedents was 39 years (range 0-94

years). When compared to the New Mexico population, White non-Hispanics and American Indian decedents are over-represented among drowning deaths. Drowning deaths most commonly occurred in lakes, followed by bathtubs, and then rivers and ditches, though this varied by age group. For example, the majority of toddler (18 months-3 years) deaths occurred in swimming pools. May through August captured the majority of incidents. Nearly half of the decedents had drugs or alcohol present at the time of death. In terms of manner of death, most (90%) were ruled to be accidental; 2% homicide, 4% suicide, and 5% undetermined. Most common findings at autopsy were pulmonary edema, skin maceration, hyperinflated lungs, and water in sinuses; only pulmonary edema was present in the majority of cases (80%).

In conclusion, having less water present does not decrease the risk of drowning deaths. Similar to national trends, males are over-represented in drowning deaths. Irrigation ditches (arroyos) as significant potential drowning sites may be a feature unique to desert states. Public health strategies aimed toward increasing awareness of drowning potential, importance of sobriety while in/around water, and expanding availability and popularity of swimming lessons may help to decrease the disturbingly high rates of drowning deaths in New Mexico.

Reference:

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Drowning, Desert, Public Health

G101 Reye's Syndrome: A Proposal of Two Fatal Cases

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The goal of this presentation is to examine the clinical and histopathological aspects of two cases of Reye's Syndrome.

This presentation will impact the forensic science community by describing the necessity of a complete methodological forensic approach by means of autopsy and histopathological examinations to diagnose Reye's Syndrome. The anamnestic element, like this case shows, can be fundamental to direct the diagnosis in the correct way.

Reye's Syndrome (RS) is a descriptive term covering a group of heterogeneous disorders. Various factors were considered in the pathogenesis of RS, such as viral, toxic, drug-related, and metabolic. The link between aspirin ingestion and RS has not been scientifically demonstrated. Neither has the link between salicylate ingestion and a range of inherited metabolic disorders that can mimic RS (Reye-like diseases). Epidemiological data seems to show a significant reduction in the incidence of RS after the Centers for Disease Control and Prevention (CDC) issued warnings in the U.S. in 1980 against aspirin administration to children.

The first signs of RS are usually persistent vomiting and diarrhea. Other early symptoms may include irregular breathing, listlessness, drowsiness, lethargy, and coma. Fever is not usually present. The causes of symptoms associated with RS relate to dysfunction of the liver and a resultant increase in serum ammonia levels and other toxins. These toxins cause increased pressure

in the brain and swelling, leading to brain dysfunction, and can progress to death.

Case 1: A 10-month-old girl was admitted to the hospital for a deep coma state (Glasgow Coma Scale (GCS) 3) during acute bronchitis and persistent fever. Her temperature was 39.7°C, her pulse was 125 beats per minute, and she was noted to be gasping. Her parents revealed they gave aspirin to their daughter for about four days prior to admission because of the flu. There was no previous history suggestive of a metabolic disorder.

A chest Computerized Tomography (CT) scan demonstrated the presence of pulmonary edema and incomplete flow of lower lobes. A brain CT scan showed diffuse, hypoxic-ischemic changes and slight cytotoxic edema of the parenchyma.

Chemical analysis showed elevated Serum Glutamic-Oxaloacetic Transaminase-Serum Glutamic Pyruvic Transaminase (SGOT-SGPT) without jaundice, elevated blood ammonia level, metabolic and respiratory acidosis (pH 7), hypoglycemia, and coagulopathy (PT%: 66.5, PT sec: 13.56). The child died ten days after admission. The autopsy revealed hepatomegaly and diffuse brain edema with massive bronchopneumonia. The etiopathogenetic definition was outlined by histological examinations of all organ samples, using Haematoxylin and Eosin (H&E) and immunohistochemical staining methods that revealed the presence of a diffuse microvesicular accumulation of fat in hepatocytes, diffuse vasospastic cerebral edema with small endovascular hemorrhages, myocardial myocytolysis, and widespread stasis of all the remaining organs. The death was attributed to multi-organ failure.

Case 2: A 12-year-old female presented with fever, asthenia, lack of appetite, and diarrhea for a week. Aspirin was administered daily. Seven days later, the patient suddenly lost consciousness in bed. On admission to the hospital, the child was in a deep coma with fixed mydriasis, areflexia, absence of response to any stimulus, and decerebrate posturing. Blood chemistry showed high SGOT (1555U/L) and SGPT (1395U/L), hypoglycemia (10mg/dL), and elevated prothrombin time (130sec). She died a few hours after admission. The HbSAg test was negative. The autopsy revealed yellow complexion due to jaundice, brain edema, subserosal petechiae, lung congestion and signs of hemorrhages, intensely yellow liver with creamy consistency, and diffuse subcapsular hemorrhages. The etiopathogenetic definition was outlined by histological examinations of all organ samples, using H&E and immunohistochemical staining methods that confirmed stasis of all organs and diffuse hemorrhagic foci. The liver showed typical features of massive centrilobular necrosis and vacuolar degeneration of liver cells. Death was attributed to RS.

In conclusion, RS should be suspected when this pattern of symptoms appear during or, most commonly, after a viral illness with (or with out?) aspirin ingestion prior to presentation. Not all of the symptoms have to occur, nor do they have to be displayed in this order. Many diseases have symptoms in common and a complete autopsy with histopathological examination can aid in the correct determination of the cause of death.

Reye's Syndrome, Liver Failure, Cerebral Edema

G102 Skin Artifacts Due to Postmortem Damage Caused by *Notiothauma Reedi* McLachlan (Mecoptera: Eomeropidae): A New Insect of Forensic Importance in Forests of Southern Chile

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After attending this presentation, attendees will be knowledgeable about a new biological marker for decomposition of mammal carcasses in rainy forests of southern Chile, South America.

This presentation will impact the forensic science community by showing that postmortem artifacts caused by insects found in carcasses could be important to consider in a forensic analysis.

The phenomenon of postmortem animal interference to human bodies is part of the taphonomic processes of corpses, occurring in the early and late part of the postmortem period. Adequate training in forensic pathology requires familiarity with the types of artifacts that may be introduced by predatory animals and, when evidence of animal activity is found at autopsy, consideration of a wide range of possibilities is required. It is important to consider that postmortem animal feeding activity may cause considerable damage to bodies resulting in the modification of wounds, loss of identifying features, and injury or removal of internal organs. Certain postmortem lesions may appear inflicted as antemortem injuries, with consequent potential problems in investigation interpretation. This study presents the first report of postmortem skin artifacts in pig carcasses caused by *Notiothauma reedi* (Mecoptera: Eomeropidae), and discusses the implications of this species being a new bioindicator in the forensic entomology in Chilean Valdivian forests.

This investigation will show to the public that postmortem artifacts caused by insects found in the carcasses could be important to consider in a forensic analysis. The experiment was designed to describe and compare the succession of insects associated with the process of decomposition of the carcasses and was performed in the winter of 2013 simultaneously in five distinct areas of La Araucanía region, Chile: (1) second-growth native evergreen forests; (2) native deciduous forests; (3) a plantation of *Eucalyptus nitens*; (4) a plantation of *Pinus radiata*; and, (5) an anthropized prairie. Thirty 20kg pigs (*Sus scrofa Linnaeus*) were used, being deposited on site by means of encephalo-cranial trauma. The carcasses were placed in a left-lateral decubitus position inside cages consisting of soldered bars covered by metallic mesh with dimensions of 100cm x 80cm x 60cm to avoid the intervention of vertebrates and to allow easy access for entomofauna. The following parameters were recorded daily: (1) environmental temperature; (2) rectal temperature of the carcasses; and, (3) rainfall and environment humidity.

On day 3 of the Postmortem Interval (PMI), the carcasses entered the emphysematous stage, and the first adults of *N. Reedi* were found feeding on the skin of the carcasses located in all the studied forested environments, but were not found in the anthropized prairie. The areas of the skin damaged by *N. Reedi* were characterized as round lesions, about 3-5 millimetres in diameter, with an alopecic border. The slightly eroded centers of the lesions were reddish-brown and involved the epidermis and part of the superficial dermis. Interestingly, it was not observed that *N. Reedi* makes deep lesions in the skin, but rather eats part of the skin and then goes on to do the same in another area, making these characteristic lesions. In advanced stages of the decomposition process (day 45 PMI), the edges of the lesion are raised with fungal colonies. In this state of decomposition, the skin color is greenish with abundant yeast and hyphae of other fungus; however, in the peripheral area of the lesion the color is red and in the central area of the eroded lesion the color is dark red. The morphological characteristics of postmortem artifacts observed and described in the skin of the pig carcasses have similarities in

size, shape, and color with antemortem injuries in the skin caused by cigarette burns.

It is proposed by this study that the artifacts in postmortem skin caused by *N. Reedi* should be taken into consideration in the thanatological analysis of cadavers found in wooded areas where this species is present. Because this species grows only in the forest environment, it could be considered indicative of decomposition in the native forest and exotic plantations in the south of Chile.

Carcasses, *Notiothauma Reedi*, Biomarker

G103 Identification of Flesh (Carrion) Source From the Stable Isotope Analysis of Blow Fly Larvae, Pupae, and Adult Flies

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After attending this presentation, attendees will learn about the ability of stable isotope measurements to link blow fly (*Calliphora vicina* Robineau-Desvoidy) larvae, pupae, and adult flies to different carrion sources. *C. Vicina* plays a major role in carrion decomposition and corpse colonization throughout the U.S. Attendees will also gain insight into systematic changes of stable isotope ratios through the different stages of development.

This presentation will impact the forensic science community by helping to understand that the relationship between the stable isotope ratios of carrion sources, larvae, pupae, and adult flies will enable forensic entomologists and criminologists to classify or identify the food sources of immature and/or adult blow flies. This capability would enable standoff or remote species determination of decaying carrion via the collection of dispersing adult blow flies.

The variations of the carbon, nitrogen, sulfur, oxygen, and hydrogen isotopes in different objects are widely used by forensic chemists to prove the authenticity of the objects, to find their origins, or to discriminate between endogenous and exogenous compounds in drug abuse cases. Because the isotope ratios of carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) are good representatives of a food source, they are frequently used in criminology to predict the previous geographic locations of an unidentified body. Ecologists often use isotope ratio analysis to determine the trophic level of organisms and their primary food sources. However, such analyses are rarely interested in linking adult insects to a specific meat source in a forensic context. This study begins with a proof-of-concept study to test the hypothesis that immature and/or adult blow flies can be linked to specific food sources via their stable isotope ratios.

Adult flies of a commercial strain of blow fly (*Calliphora vicina* Robineau-Desvoidy, Diptera: *Calliphoridae*) were allowed to oviposit on raw pork muscle, raw beef muscle, raw chicken liver, or a 20% human blood agar. The eggs were allowed to hatch, feed, and develop into adult flies. Ten individuals from each meat source were arbitrarily selected at the following stages: postfeeding third instar; pupa and associated puparium, and newly-emerged unfed adult flies. The bulk $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values for the different stadia were established using an Elemental Analyzer-Isotope Ratio Mass Spectrometer (EA-IRMS). The differences between the isotope ratios for each life-cycle stage were determined by 1-way Analysis of Variance (ANOVA) and post-hoc pairwise comparisons.

In most cases, the bulk $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ isotope ratios were significantly different between the larvae, pupae, and adult flies for every type of meat source, indicating a significant degree of isotope fractionation at each stage of development. The $\delta^{13}\text{C}$

values for the pupae from the pork and chicken samples were the only exceptions. Although fractionation measurable, the extent of fractionation was typically an order of magnitude smaller than the initial differences between the original meat sources, indicating that it is indeed possible to link each meat source directly to each stage of *C. Vicina* life cycle. Due to the multiple biological processes in the organisms throughout their development, the bulk $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values for the larvae, pupae, and flies differ significantly from the initial meat source, but the changes in the $\delta^{15}\text{N}$ values throughout the biological development stages had the same trend for each carrion type, which can eventually lead back to the original meat source. The planned Compound-Specific $\delta^{13}\text{C}$ Analysis (CSIA) of larvae, pupae, and flies will give additional information about the relationship of the flesh type and the species grown on them. Furthermore, CSIA will allow elucidation of the amino-acids which are most representative of the initial food source.

Isotope Ratios, Flesh Source, Blow Flies

G104 Bioinformatics Tools and R-Language Programming for the Classification of Soils

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After attending this presentation, attendees will learn and better understand some of the principals of soil forensics. There are vast amounts of complex data that can be collected from soil samples and, by using bioinformatics tools and the R-language programming, it is possible to deconvolve these complexities and establish classification schemes for soil that can assist in its provenance.

This presentation will impact the forensic science community through the implementation of the methods and analyses described. Bioinformatics tools, once learned and understood, provide a simple and rapid means of analyzing large datasets, such as those obtained from soil samples. The potential of this study also lies in the creation of a searchable database for soil forensic identification.

Soil is a highly dynamic substrate consisting of many physical and chemical properties as well as a rich array of biological diversity. These soil properties make it a very useful and informative type of evidence for forensic investigators if all of this information can be analyzed. The ecological hypothesis states that soil type, which is characterized by the physical and chemical properties of the soil, is also highly correlated to the soil biota and microbial habitat.¹ Therefore, soil metagenomic profiling can provide a rapid tool for the discrimination and classification of soil. Previous studies have demonstrated the usefulness of physical and chemical properties of soil for accurate classification using elemental analysis but it was shown that 16SrRNA biotic profiling was more effective.² Another soil study applied bioinformatics tools such as support vector machines and K-nearest neighbors to distinguish soil bacterial community-pattern differences in soils from Idaho using the hypervariable 16SrRNA domains. These methods were able to predict the classification of soil (location and/or treatment) from the microbial profiles with high accuracy.³

Machine-learning tools are widely used for classification purposes and there are both supervised and unsupervised methods. These methods train on a known data set to recognize patterns; subsequently, unknown samples can be tested against the training set and classified as to their similarity. The current study used physical and chemical properties of ~1,270 soil samples from

various geographic locations across Miami-Dade County, Florida. Biotic profiles were generated by Polymerase Chain Reaction (PCR) from the taxonomic soil groups — bacteria, archaea, fungi, and plant — for each soil sample. The combined dataset of chemical/physical properties and microbial profile data were used to train the machine-learning algorithms that were programmed using R. In particular, random forest, decision trees, and neural networks were implemented and compared for accuracy of classification. When using abiotic and biotic data separately, the classification accuracy was not as high compared to when both were concatenated. This was true for both random forests and decision trees but, curiously, not of neural networks. Random forest analysis, when combining both biotic and abiotic data, was able to classify “test” samples to their known soil type with 100% accuracy; decision tree analysis had a 98% accuracy rate. Neural networks were unable to classify soils accurately when both biotic and abiotic data were combined, with a low rate of 35%. Although, when tested separately, both abiotic and biotic data were able to classify soils with good accuracy, using neural networks. This approach demonstrated that the use of both chemical/physical properties as well as microbial community patterns can provide higher accuracy in the discrimination and classification of soils and provides a rapid method of analysis for forensic investigators to determine soil provenance. Future work will involve the implementation of K-nearest neighbor and support vector machine algorithms to add to the suite of learning tools for a more comprehensive comparison of their usefulness for soil classification.

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Soil Forensics, Machine Learning, Microbial Profiling

G105 Impact of the Estimation of the Body Weight on the Determination of the Postmortem Interval

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After attending this presentation, attendees will know the forensic pathologist's ability to estimate the weight of a corpse and how the knowledge of the body height influences his estimation.

This presentation will impact the forensic science community by raising awareness of the impact of an over- or under-estimation of the body weight on the determination of the PMI using standard methods.

Background: The forensic pathologist has to do his/her

best to estimate as precisely as possible the Postmortem Interval (PMI) when called upon to examine a dead body at the scene. Different methods based on rectal temperature exist to help the pathologist with this issue, like the Henssge nomogram or the Marshall model. However, the body weight is required to use these methods, because the rate of body cooling depends greatly on it. This implies that the weight is known, or is at least estimated, at the scene.

Objectives: This study intends to assess the forensic pathologist's ability to estimate the weight of a corpse, to determine if the knowledge of the body height improves his estimation, and to show how such an estimation impacts the determination of the PMI.

Methods: A total of 50 corpses subjected to an autopsy were selected in the Forensic Department of the University Hospital of Montpellier regardless of their age, sex, and state of preservation. Their weight and height were measured before autopsy was performed, but they were not initially disclosed. The weight of each corpse was estimated in a totally blind manner by three evaluators composed of two forensic pathologists and a house physician. The evaluators had to estimate the weight of each corpse before and after being aware of the body height, so they were able to change their initial estimation. The approximate body weights and the actual body weights were reported. From these figures, PMIs were calculated, then compared in each case, using both methods mentioned above. Various statistical tests were used. Constant predetermined values were entered for the variables taken into account in these methods (ambient and rectal temperatures, correction factors), excepted for the height which had been measured.

Conclusion: The results of this study will reveal how accurately a forensic pathologist can estimate the weight of a corpse, how the knowledge of the body length may influence that estimation, and the impact the over- or under-estimation of body weight has on the determination of the PMI, using standard methods.

Postmortem Interval, Body Weight, Estimation

G106 Age Estimation Using T-Cell Receptor Excision Circles (TRECs) in Tissue Samples for Forensics

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After attending this presentation, attendees will understand the basic principle of T-Cell Receptor Excision Circles (TRECs) formation, applicability of the method for other types of tissues, and characteristics of quantifying TREC patterns in each tissue.

This presentation will impact the forensic science community by providing the possibility of age estimation using tissue samples.

Age estimation using biological remains is one of the interesting topics in forensics. Various approaches have been performed to estimate age, and quantifying DNA molecules existing in T cells is one of the recent promising methods. However, quantification of that molecule has only been performed in blood samples; it has never been tried in any other type of human tissue, nor has its applicability in forensics been verified.

The central role of the thymus in T-cell production and T-Cell Receptor's (TCR's) gene rearrangement is well established, together with thymic changes with time (i.e., thymic involution).

During the rearrangement of TCR gene segments, some regions, which were not selected to form parts of TCRs, are spliced out as circular DNA. One of these DNA is a signal-joint T-cell Receptor Excision Circle (sjTREC), which exists in naïve T-cells which have undergone development and maturation in the thymus. SjTRECs do not replicate during cellular proliferation, but rather are diluted by each round of cell division. Therefore, it is supposed that the content of these episomal DNA per total number of T-cells would be decreased with aging. In forensics, the application of these molecules for estimating age is being investigated.

In some forensic cases, the condition of blood samples would be not applicable to quantify sjTREC contents. For example, blood obtained from decomposed bodies could be putrefactive or even non-existent, since the decomposition of tissues containing a larger quantity of water would be processed earlier than that of other tissues. The investigation of alternative body samples could be helpful to such cases. In this report, the thymus and spleen were selected as potential samples, as they are the primary and secondary lymphoid organs, respectively. It is thought that these tissues are closely related with thymopoiesis and T-cell homing.

Three tissues, blood, thymus, and spleen, were obtained from dead bodies in the age range of 4-70 years through autopsies, and genomic DNA was extracted. The measurement of sjTREC contents was performed using real-time TaqMan® Polymerase Chain Reaction (PCR), since it is essential to detect sjTREC in the peripheral blood precisely and sensitively. The probe is sequence-specific and primers allow only excised-out sjTREC DNA from TCR gene rearrangement to amplify.

The quantification of sjTREC molecules was possible and declining trends with aging were confirmed in the samples extracted from both the thymus and spleen. The pattern of sjTREC levels with age in each tissue was not identical, and its level of samples from blood, thymus, and spleen showed a different pattern in one individual. Further research is required to understand the meaning of or reasons for this difference.

Age Estimation, Thymus, TRECs

G107 The “Overkill” Gang Homicides

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The goal of this presentation is to present multiple murders carried out by a youth gang in the most violent manner with much cruelty. A detailed crime scene investigation and a complete postmortem examination established the different contemporary weapons used for the killings.

This presentation will impact the forensic science community by emphasizing the problem of serial murders committed by emerging young gangs which, although widely present and frequent in the U.S. and Latin American countries, are not frequent or that common in Italy.

Gang homicides involving the use of different injurious means at the same time are rarely reported in the literature. Often, these gang-related murders occur with firearms and in public places, suggesting that gang homicides are quick, retaliatory reactions to ongoing gang-related conflicts. In fact, the homicides committed in southern Italy are principally attributable to groups of crimes organized locally, which control several illicit activities and are related to continual ongoing fights among the members of the local gangs. These crimes are typically committed to enact

revenge, challenge reasons related to robbery, theft, settling, drug and mental disorders.

This study introduces four cases of homicides, committed in a northern town of Puglia, between June and October of 2012, by a band of boys between the ages of 17 and 31 years and comprised of six men and one woman.

The first two murders, committed at the same time, were related to drugs. The two victims were found just outside the city. One was in his car and had two gunshot wounds in the skull. The other was found in a field not far from the site of the murder with burned clothes and skin. Three gunshot wounds were detected in his head and thorax. The autopsy showed that he was killed by blows from a firearm and was later burned.

The third murder, committed for futile reasons, involved a 21-year-old male who disappeared in July 2012. His remains were found in December 2012 in a farmland's field. Skeletal remains as well as fragments of muscular tissue, still wearing clothes, were completely buried under the mud in a prone position with the legs inside a plastic bag. In the field near the dead body, a bracelet, a piercing, and four bullets were found. Forensic activities involved external examination and radiological investigation by means of total body multislice Computed Tomography (CT) scan. Examination of the clothes helped to determine the different tools used to kill him, which were a firearm, blunt force, and a steel weapon, variously combined. The bones of the skull showed two gunshot wounds and many scratches from a blunt instrument. Several vertebral fractures and stab wounds on a rib and on a shoulder blade were recorded. Five anterior and six posterior tears on the right sleeve were described on the t-shirt; three tears were also found on the left leg of the shorts.

The last murder involves a 60-year-old man, killed for money reasons. The man was found dead in his own garage, lying prone on the floor. The floor was covered in blood close to the head. His wrists and ankles were bound by tape, and his neck and arms were tied together at the chest. At external examination, some incised wounds with regular margins were described on the neck and multiple linear lacerations with irregular and bruised margins were noted on the head. At gross examination, galea capitis showed massive hemorrhagic infiltration. Multiple linear fractures of the skull vault were detected. The neck revealed massive hemorrhages in the subcutaneous tissues and muscles and the right jugular vein was sectioned.

As established by other international publications, the use of drugs and violence are behind these gang-related murderers. The different tools and weapons which were used (firearms, blunt force, and “cutting” weapons) and the high numbers of injuries seen on the victims' bodies are a clear and unquestionable sign of the cruelty of these crimes.

Gang Homicides, Extreme Violence (Overkill), Different Weapons

G108 Comparison of Three Novel Polymers Using Capillary Electrophoresis for Bioseparations of Complex DNA Mixtures

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After attending this presentation, attendees will understand the basics of electrophoresis and important parameters that influence DNA separation in Capillary Electrophoresis (CE). Polymers are the most critical parameter that influences CE resolution by altering DNA migration behavior.¹ The most common polymer for CE analysis is POP-4; however, it is unable to separate

same-length amplicons that contain a different base composition.² Therefore, this study will demonstrate to the attendees the novel polymers that have the potential to resolve this dilemma.

This presentation will impact the forensic science community by demonstrating novel polymers that can provide 2D mixture profiles based on length and sequence differences. These novel polymers have the potential to assist in the detection and identification of harmful pathogens and biothreat agents that are critical to homeland security as well as be applied to soil forensics by utilizing microbial profiling of soils as collaborative evidence. Currently, this is difficult to do with traditional techniques due to soils' complex biological and physical properties.³

Microbial communities are diverse arrays of organisms that have complex interactions, genes, and gene functions.⁴ There are different levels of resolution commonly used to study microbial communities. Currently, DNA sequencing/metagenomic analyses are the highest resolution used to characterize communities. However, not every analysis needs that depth of resolution, so often community profiling via amplicon length sequence heterogeneity is employed. However, the diversity is grossly underestimated because analyses are based on the number of bases in the amplicon versus the sequence polymorphisms. Taxonomically unrelated organisms can produce the same-length amplicon but have different nucleotide sequences.² A critical need exists to develop a method that can rapidly analyze community profiles not only by length, but also based on inherent sequence polymorphisms without the need for metagenomic sequencing. The commercial polymer (POP-4) and three novel polymers (F-108, PVP/HEC, and G-gels) were compared using an ABI® Genetic analyzer CE to discover the best matrix for separating and detecting the obscured sequence diversity within length-based amplicons of microbial populations.^{1,2,5} Four model organisms that display the same-length amplicon for hypervariable domain V3 within the 16S rRNA gene, but have variable nucleotide content within the amplicon were amplified by Polymerase Chain Reaction (PCR) using 16S rRNA universal primers and separated by capillary electrophoresis. More complex systems were then analyzed by artificially mixing the four isolate's DNA and ultimately analyzing a complex natural community, cyanobacteria-dominated microbial mat from Hunter's Hot Springs in Lakeview, Oregon. F-108 polymer displayed the best results, showing all four amplicons. Moreover, F-108 did not underestimate the true diversity of the microbial mat community. G-gels could not be reproduced in the ABI® CE 310 following the published parameters.² Combined G-gel with POP-4 and PVP/HEC illustrated similar results to commercial POP-4; therefore, they were not able to separate same-length amplicons with differing base composition.

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Capillary Electrophoresis, DNA Separation, Polymers

G109 Unusual Case of Asphyxia Due to an Excessive Use of Denture Glue

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After attending this presentation, attendees will be aware of the possibility that denture glue can be dangerous and, especially in people with mental disorders, can cause asphyxia and death.

This presentation will impact the forensic science community by providing an unusual case of asphyxial death by aspiration of denture glue, which is very uncommon because the denture glue is safe if properly used.

In March 2013, a 60-year-old woman was found unconscious, supine in the bathroom of her house by her son. He called for help and started cardiopulmonary resuscitation. Physicians arrived about 15 minutes later; however, the cardiopulmonary resuscitation attempts were unsuccessful and she was pronounced dead at the scene. Physicians noticed that a large amount of white sticky material was in the mouth. They also found the upper denture plate, which was then removed. The woman's husband reported that she used an excessive amount of denture glue and that she suffered from psychiatric disorders, particularly depression, so she was in therapy with antidepressant and antipsychotic drugs.

The autopsy was performed the day after the body was found. The decedent was a 60-year-old woman. At the time of the autopsy, the well-preserved body was found to be 160cm in length and weighed 55kg. On the external examination of the corpse, no macroscopic evidence of trauma was found. Abundant whitish sticky material completely filled the mouth and flowed out. At the autopsy examination, the same material was found in the pharynx, larynx, esophagus, and stomach, filling them completely and almost reproducing their shape. The denture glue obstructed the larynx, clogging the passage of air into the lungs. Pulmonary edema and congestion was also found. Toxicological exams were performed on samples of blood, bile, urine, and gastric contents, but they only showed therapeutic levels of the antidepressant and antipsychotic drugs that she was taking. Histological examination was performed which confirmed the pulmonary edema and congestion.

After the macroscopic and microscopic analysis, the cause of death was identified as acute mechanical asphyxia, caused by massive aspiration and ingestion of denture glue. This cause of death is also compatible with the circumstantial data. In fact, the woman's husband reported that she suffered from mental disorders and she used a lot of denture glue to prevent detachment of the denture. This case is remarkable because the use of denture glue is generally safe and no other cases of asphyxial death caused by aspiration of denture glue are reported in the literature.

Denture Glue, Asphyxia, Airways Obstruction

G110 Cremation Weight: Sex and Age Variation in North Carolina

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After attending this presentation, attendees will better understand the utility of cremains weight for the estimation of biological profile parameters such as sex. This presentation will illustrate the necessity for a better understanding of the limitations faced by forensic scientists when analyzing cremated human skeletal remains.

This presentation will impact the forensic science community by developing standards for cremation weights in North Carolina.

As commercial cremation is becoming more widely chosen over burial, this presentation will impact the forensic community by increasing the regional data available to the forensic investigator.

The Cremation Association of North America (CANA) projected that by 2010 the number of people choosing cremation over burial would increase to close to 36%. This increase in commercial cremation will most likely increase litigation involving issues of identity and negligent cremation practices such as the high-profile case of the Tri-State Crematory in Georgia.¹

Cremation weights are important sources of data in that they can be used to estimate the minimum number of individuals present in situations where commingling may be a possibility.² Regional mean cremation weights have been established for males and females for Florida, Tennessee, and California.³⁻⁵ The purpose of this study was to develop standards for North Carolina using a large sample of unclaimed cremains held at the North Carolina Office of the Chief Medical Examiner (NC OCME) from the years 2005 to 2013. The sample totals 303 (♀ = 60; ♂ = 243) individuals with known demographics (e.g., age-at-death, forensic weight, biological sex, and stature). The cremains are stored in a plastic urn weighing 0.5kg. Cremains were weighed using an autopsy digital floor scale and the box weight was accounted for by calibrating the scale to include the weight of the urn. Interestingly, the mean weights for males and females are greater than published weights (mean ♀ weight=2,766.96g, SD=623.29; mean ♂ weight=3,531.73g, SD=651.27). Results show that male and female cremains weights are significantly different ($F(1, 301) = 67.46, p < 0.0001$). A logistic regression was conducted to examine sex classification parameters. The following equation can be used to estimate sex from an unknown set of cremains: $\text{Sex} = 4.5938037 + -0.0019079 * (\text{cremains weight}), \text{SE} = 0.8694189$. A positive value would indicate a female, while a negative value would indicate a male. Individuals were also grouped into the following age categories (1 = 19-30, 2 = 31-50, 3 = 51-65, 4 = 66+) in order to examine if age had an effect on cremains weight. This was examined with a one-way Analysis of Variance (ANOVA) conducted on each sex separately. Results show that age-at-death does not have an effect on cremains weight for either females ($F(3, 55) = 2.69, p = 0.06$) or males ($F(3, 237) = 2.31, p = 0.08$).

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Cremains, Sex, North Carolina

G111 Making the Cut: Patterns of Association Between Victims, Suspects, and Body Treatment in Postmortem Dismemberment Cases

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After attending this presentation, attendees will appreciate how the characteristics of homicide cases involving postmortem dismemberment of the corpse differ according to the sex and age of both the victim and the suspect as well as how patterns derived from a larger sample of such cases compare to a series of ten dismemberment cases from Maricopa County, Arizona.

This presentation will impact the forensic science community by providing data that suggests that the demographics of the victims in dismemberment cases as well as how the body was treated may be associated with certain characteristics of their killers. This information has potential utility both in generating and prioritizing lists of potential suspects.

Published accounts of 94 homicide cases involving postmortem dismemberment were taken from the literature and information regarding the age and sex of both the victim and the suspect, the relationship of the victim to the suspect, the cause(s) of death, the minimum number of body segments produced by the dismemberment, and the manner of disposal of the body were recorded and compiled into a database. Where data were sufficient to permit statistical analysis, such analyses were carried out with a significance level of $\alpha = 0.1$. Patterns were sought between variables, especially as related to the age and sex of both the victim and the suspect, and any patterns that were identified were then compared to a series of ten dismemberment cases from Maricopa County in an effort to see if these patterns held when applied to novel cases.

Results from the compiled database suggest that male and female victims differ significantly in terms of the relationship that they held with their killer (Fisher's Exact Test, p -value = 0.0001204). Female victims were more frequently killed by people with whom they had an intimate relationship and males were more often killed by acquaintances. Male and female victims also significantly differed in terms of their causes of death (Fisher's Exact Test, p -value = 0.07624), with strangulation a more frequent cause of death for female victims, and some variety of sharp force trauma more frequent among males. Although missing data within the published accounts preclude statistical verification, the data also suggest that female victims tend to be sectioned into more pieces than male victims, that a body sectioned into more than six portions is much more frequently associated with a male suspect, that female suspects tend to kill intimate relations, that strangulation is more frequently employed by suspects younger than 35 years of age, and that older suspects tend to section decedents into a larger number of pieces than younger suspects.

A series of ten dismemberment cases from Maricopa County, Arizona, were used as a comparative sample to evaluate the patterns identified above. Contrary to the trend identified in

the compiled database, only one female victim from Maricopa was killed by an intimate. In four of the six cases involving male victims, the suspects were acquaintances of the victim, which generally supports the pattern noted above. Differences in the cause of death as described above are not evident in the sample of cases from Maricopa. Likewise, the associations of strangulation with younger suspects and hyper-sectioning of the corpse with male suspects are absent in this sample. However, the remainder of the trends noted above is generally borne out within the Maricopa County cases.

Although rigorous comparisons were rendered impossible due to missing and inconsistently reported data, a number of trends that relate victim characteristics and suspect characteristics were identified and evaluated. These results suggest that, despite differences which might be attributed to regional variations (homicides involving gunshot wounds, for example, were only observed in North American cases), there might be underlying commonalities between homicide cases involving postmortem dismemberment. Some of these commonalities, such as the association of strangulation with younger suspects or that of a large number of cuts with male suspects, might potentially be useful in either generating or prioritizing suspect lists. More complete data are required in order to ascertain whether or not these patterns can be statistically verified; however, the associations identified in this preliminary research provide a guide when analyzing dismemberment cases in a forensic setting.

Postmortem Dismemberment, Victim/Suspect Relationship, Dismemberment Patterns

G112 A Case of Reconstruction of Carbon Monoxide Intoxication From Burning Ignition Charcoal Briquettes to Confirm a Suicide

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After attending this presentation, attendees will learn the importance of collecting the components of Ignition Charcoal Briquettes (ICBs) from hands and nostrils to link the deceased and ICB to prove the Carbon Monoxide (CO) intoxication to be a suicide and also learn the analytical techniques for the analyses of ICB, CO, diphenhydramine, and polymer resin.

This presentation will impact the forensic science community by providing suitable methods to carry out when there is an intoxication suspected of being CO.

ICBs, used for igniting briquettes and normally consisting of blackened sawdust with barium nitrate ($Ba(NO_3)_2$), are frequently employed to commit suicide inside automobiles or sealed rooms. CO intoxication produced by burning ICBs is a primary method of suicide in South Korea. However, some CO intoxications could be classified as homicides so it is crucial to prove that the connection between the deceased and the event scene be established by analyzing the evidence collected from the scene to determine if it was intended to be a suicide or homicide.

In this case, a man was found dead on a bed in a motel room with a burned charcoal briquette in a metal container about 2 meters away from the bed. This study employed a variety of evidence to connect the ICBs and the deceased to clarify the event and identify the cause of death. Approximately 30 pieces of evidence were submitted by investigators to identify the link between the deceased and the door locks of the motel room to recreate the deceased's path before the incident. Samples were

also collected from the nostrils to predict if the deceased was close to the ICB when it was ignited. The blood was tested for CO-Hb concentration and medications, particularly sedative drugs. A partially burned resin-type material was collected to determine what it was used for. A stereomicroscope, a microscope Fourier Transform Infrared Spectroscopy (FTIR), and a Scanning Electron Microscope-Energy Dispersive X-Ray Spectrometer (SEM-EDX) were employed for the analysis and comparison of components of a charcoal briquette. Also used were a Gas Chromatography-Thermal Conductivity Detector (GC/TCD) to detect the CO-Hb level, a Gas Chromatography-Mass Spectrometry (GC/MS) for sedative drugs, a stereomicroscope, a microscope FTIR, and a pyrolysis-GC/MS for a burned resin-type substance.

ICBs are round-shaped (about 14cm in diameter and 5cm thick) with holes in them and are pasted with $Ba(NO_3)_2$ on one side of the center surface. Setting a fire on the side with the $Ba(NO_3)_2$, flame abruptly starts and some portions of the ICB can be transferred to surrounding surfaces, including the nostrils. This study confirmed the components of ICB on the locks of the motel room and hands of the deceased by identifying black color particles morphologically with the stereomicroscope, microscope FTIR (confirming no special organic absorption peaks), and SEM-EDX by obtaining carbon as a main element, which meant the deceased might have transported the ICB. Black material found in the nostrils, consisting of carbon and barium elements, would have been inhaled as the ICB emits these elements as soon as they are ignited. This indicates that the deceased was near the ICB and it can be assumed that he set the fire.

A carboxyhemoglobin level of 79% was obtained, indicating the cause of death was intoxication of CO. In addition, 2.3mg/L of diphenhydramine, a sleeping drug, was detected, which is lower than a fatal dose (8-31mg/L (mean 16mg/L)).¹ The partially burned material was analyzed as an acrylonitrile-styrene polymer, normally used for making shopping bags, which might have been used to carry the ICBs.

In this case of reconstruction for CO intoxication, the following statements were able to be determined after scene investigation, autopsy, and laboratory testing: confirmation of who had carried the ICB to the motel room was confirmed by identifying the components of the ICB on the locks of the room and hands of the deceased; the igniter was determined by confirming the components of the ICB, primarily carbon and barium elements, in the nostrils of the deceased; the cause of death was verified by analyzing the CO-Hb level in blood which was 79% (a fatal level); the deceased was found to have taken a sleeping pill (diphenhydramine) with a non-fatal level of the drug; and an acrylonitrile-styrene material bag was used to carry the ICBs. Serial steps mentioned above could be a paragon to draw the conclusion of suicide by intoxication of CO.

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Carbon Monoxide Intoxication, Ignition Charcoal Briquettes, Suicide

G113 Fatal Death by Poisoning: From Myth to Science — A Forensic Point of View

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After attending this presentation, attendees will understand the prevalence of poisoning from the forensic science perspective.

This presentation will impact the forensic science community by demonstrating the forensic and anthropological aspects of poisoning throughout the review of literature.

Introduction: Toxicology is "the science of poisons." More specifically, the chemical and physical properties of poisons, their physiological or behavioral effects on living organisms, the qualitative and quantitative methods for their analysis, and the development of procedures for the treatment of poisoning constitute the field of toxicology. Although the history of poisons dates to the earliest times, the study and the science of toxicology can be traced to Paracelsus (1493–1541) and Orfila (1757–1853). Poisoning is a major public health problem worldwide and a leading cause of injury and/or death in the United States. Ambade et al. demonstrated that in suicidal deaths, poisoning (42.3%) was the most common method of suicide followed by burning (21.5%). Moreover, mortality due to unintentional drug poisoning increased by 62% from 1999 to 2004 in the U.S. The largest increase in unintentional poisoning mortality was observed among adults aged 20–29 years and 45–54 years, women, White, or populations living mostly in rural states. Analysis of the literature was performed through the standard search engines, and in particular PubMed NCBI. The studies were divided according to the substance being analyzed. Subsequently, the tables of the collected data were carried out and the studies were divided according to the manner of death and the method of administration of the poison. The scientific articles were divided into three groups.

Conclusions: It can be concluded that the greatest number of deaths from accidental cause was found in group I (animals, plants, and fungi), while the highest number of deaths due to suicide was found in group II (herbicides, pesticides, rat poison). In group III, the situation is similar to both the deaths due to accidental cause and suicide. Poisoning is no longer considered the primary choice to hide a murder, contrary to what is told in myths and what actually happened in past centuries.

Poisoning, Forensic Science, Manner of Death

G114 Sudden Death Due to Ruptured Pseudoaneurysm of Femoral Artery in Injected Drug Abusers: A Case Series

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After attending this presentation, attendees will understand some principles of an emerging as well as a serious complication of pseudoaneurysm in injected drug abusers.

This presentation will impact the forensic science

community by making attendees aware of an emerging problem in injected drug abusers, as most of the reported cases of pseudoaneurysm found in scientific literature are secondary to iatrogenic intervention. The presentation will highlight the case history and autopsy findings of these cases with relevant photographs of this catastrophic event in injected drug abusers.

Substance or drug abuse has broad dimensions worldwide, particularly in the young. In the United States in 2009, its prevalence was about 8.7% among the 12-years-old-and-above. In India, cannabis, heroin, and pharmaceutical drugs are the most frequently abused drugs. Abuse with intravenous injections of analgesics such as dextropropoxyphene is reported from many states, as it is easily available at a tenth of the cost of heroin. Injected drug abuse is associated with a wide variety of manifestations. These range from bleeding from the site of injection, frequent infections, ulceration, scarring, deformity, gangrene, chronic obstructive pulmonary disease, cardiac failure, and hepatic failure/insufficiency. With increased injected drug abuse, especially intra-arterial (as with heroin and easily available pentazocine), the complications of arterial diseases become increasingly important.

Pseudoaneurysm in injected drug abusers occurs due to traumatic damage to the vessels caused by repeated injections and infections. It most commonly involves the femoral artery and requires intensive management. When medical attention is delayed, the victim is prone to massive bleeding from the ruptured artery, resulting in death. Although pseudoaneurysms in injected drug abusers are known, there is a paucity of autopsy studies on sudden deaths due to its rupture. Reported are four such cases with spontaneous rupture of pseudoaneurysm in young males who were all injectable drug abusers. Two of the victims were found dead on the roadside, one died at the hospital, and another died at his residence. The pseudoaneurysm of the femoral artery in all four cases could be grossly demonstrated at autopsy and also by histopathological examination. The duration between the terminal event of rupture of pseudoaneurysm and death in all four cases was within 24 hours, signifying the suddenness and high fatality of this condition.

Forensic scientists should be aware of this emerging problem in injected drug abusers. Most of the reported cases of pseudoaneurysm found in the scientific literature are secondary to iatrogenic intervention. Once such a pseudoaneurysm ruptures externally, there is acute vascular collapse due to the large volume of blood loss which can be avoided by early recognition and intensive management. Rehabilitation of drug abusers is also recommended.

Femoral Artery Pseudoaneurysm, Injected Drug Abuse, Sudden Death

G115 Postmortem Evidence of Methadone Cytotoxicity

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After attending this presentation, attendees will have a greater understanding of the fundamental process underlying death secondary to methadone, and possibly other opiates as well. Knowledge of these changes may be generalizable to other forms of drug-related death as well.

This presentation will impact the forensic science community by alerting them to the possibility that there may be

alternate cause of death in cases of apparent overdose.

Background: Methadone concentrations in those dying of methadone toxicity totally overlap in situations where the presence of methadone is only an incidental finding. In practice, causation may be difficult to determine. The identification of an anatomic biomarker for methadone toxicity would be a useful tool. Evidence suggests that the abuse of many different drugs can lead to the occurrence of inappropriate apoptosis. Since the neurons controlling respiration are mainly located in the rostral ventral portion of the medulla oblongata, it was hypothesized that a detailed investigation of this area might disclose an anatomic marker for methadone-related death.

Objective: This study attempted to determine if methadone induced more apoptosis in the brainstems of methadone users than in the brainstems of drug-free individuals also suffering from Sudden Cardiac Death (SCD).

Design, Setting, and Participants: A single cohort of decedents with SCD who had been autopsied at a large university hospital in Palermo, Italy, were studied. In every case, a complete autopsy with toxicology testing was performed. The brains were only examined after two weeks of formalin fixation — this allowed for easier dissection of the brainstem. Multiple blocks of tissue were prepared from the area lying immediately between the inferior and the superior colliculi. These blocks included the area of the Rostral Ventrolateral Medulla (RVLM), known to contain the nucleus solitarius, which is thought to contain most of the brain's respiratory center. There were 11 participants and five age-matched controls. The slides were read separately by two individuals, both trained in neuropathology. No attempt was made at grading the anatomic changes, the only criteria being that the apoptotic process be recognizable. This determination was based entirely on the appearance of neurons in Hematoxylin-Eosin (H&E) stained sections.

Main Outcomes and Measures: The primary outcome measure was detection of the presence or absence of neuronal apoptosis and/or necrosis within the nucleus solitarius.

Results: Cells displaying evidence of both early and advanced apoptosis, consisting primarily of nuclear condensation, nuclear fragmentation, and even nuclear absence were found. These dying neurons were admixed with other neurons displaying the features of classic ischemic necrosis with eosinophilia and nuclear fragmentation. Evidence of classic necrosis was identifiable in most of the controls, though apoptosis was not.

Study Limitations: This study did not have access to the routine immuno-staining procedures used for the identification of apoptosis, but it is believed the visual identification was accurate. Since the changes of both ischemic necrosis and apoptosis develop at different rates in different parts of the brain, it is difficult to be sure whether similar changes were or were not present elsewhere.

Conclusions and Relevance: This study shows that neurons, primarily along the tractus solitarius but occasionally in other cell nuclei (even controls), are vulnerable, presumably to direct methadone directly (via apoptosis) and indirectly methadone (via hypoxia). In instances where methadone is present in significant concentrations, but apoptotic lesions are absent, it may be reasonable to assume that methadone was not the cause of respiratory arrest, though this conclusion will require considerably more confirmation.

Methadone, Apoptosis, Biomarker

G116 Suicidal Drug Overdoses in New Mexico: Medical Examiner-Investigated Cases From 2008 to 2012

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After attending this presentation, attendees will understand the changing toxicology trends in suicide overdoses and the increasing prevalence of multi-drug overdoses.

This presentation will impact the forensic science community by emphasizing the shifting patterns of drugs present in suicidal overdoses, which have already been published in regard to accidental overdoses, in order to inform mental health treatment plans, prescribing practices, and patterns of substances analyzed at autopsy.

In order to better understand the changing toxicology trends in suicidal overdoses, a review of all suicidal drug overdoses between 2008 and 2012 from New Mexico's statewide medical examiner office was conducted. During the study period, there were 365 cases of suicidal drug overdoses, with the number of suicidal overdoses in New Mexico ranging from a low of 63 in 2009 and 2011 to a high of 83 in 2008. This represents 15.1% to 21.8% of the total number of suicides in New Mexico for the years studied. The mean age of the decedents was 48 years (range 17-85). Females were significantly older (mean age 49.1 years) than males (mean age 46.2 years) among suicidal overdoses ($p=0.048$). Between the ages of 30 and 80 years, more females committed suicide by intentional overdoses than males in every age category. Females outnumbered males by a factor of 1.6:1 (226 women, 139 men) in suicidal overdoses, but made up only 25% of all suicides. White non-Hispanics were over-represented in suicides by overdose, comprising 65.8% suicide by overdose but only 36.2% of New Mexico's population, followed by 91 (24.9%) Hispanic decedents, 25 (6.8%) American Indian, 4 (1.1%) African American, and 4 (1.1%) Asian/Pacific Islander/Unknown. Out of 106 separate substances isolated postmortem, the most commonly isolated substance, found in 79 decedents (21.6% of the total), was alcohol, followed by oxycodone (73 decedents, 20%), hydrocodone (56 decedents, 15.3%), alprazolam (50 decedents, 13.7%), acetaminophen (46 decedents, 12.6%), and diazepam (45 decedents, 12.3%). Toxicologic analysis showed 1 to 11 substances isolated, with 196 (53.7% of all cases) having three or more substances isolated. The most common class of prescription drug detected was opioids, detected in 206 (56%) of the 365 cases, followed by antidepressants in 168 cases (46%), anxiolytics in 143 cases (39%), muscle relaxants in 80 cases (22%), Over-The-Counter (OTC) pain medications in 55 cases (15.1%), and antihistamines in 54 cases (14.8%). There was a statistically significant difference ($p=0.03$) in numbers of classes of drugs present by gender, with more women having multiple classes of drugs present at the time of death. Cases in which only prescription drugs were detected (no illicit drugs or poisons detected) made up the largest category of suicidal overdose deaths (310 cases, 85%), followed by combinations of prescriptions drugs and illicit drugs (30 cases, 8%), poisons (8 cases, 2.2%), 3 cases (0.8%) of illicit drugs only, and 3 cases (0.8%) with a combination of prescription drugs and poison. There was a significant association between type of overdose (illicit, prescription, poison, other) and whether or not the overdose was attributable to a single substance ($p<0.0001$). There was also a significant association between the year of the study and the likelihood of the overdose being attributable to a single substance ($p=0.03$), with significantly more deaths attributed to multiple drugs in 2012 than in 2008 ($p=0.01$).

Eighty-eight (24%) of the prescription drug-only deaths resulted from a single drug. The most frequent prescription drugs detected from a single-drug overdose were oxycodone and quetiapine in seven cases, diphenhydramine in six cases, and amitriptyline and salicylates in five cases. Increased numbers of deaths from prescription drugs in accidental drug deaths have been examined recently in the forensic pathology literature, but research focusing on suicidal overdoses has been sparse. This research focuses on the shifting patterns of drugs present in intentional overdoses to provide adequate data in order to help inform mental health treatment plans, prescribing practices, and toxicology requests in cases of suspected suicidal overdoses.

Suicide, Overdoses, Multi-Drug

G117 Envenomation and Degranulation: A Case Report of Sudden Death Following Copperhead Snakebite and Evaluation of Toluidine Blue as a Diagnostic Tool in the Diagnosis of Kounis Syndrome

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After attending this presentation, attendees will be able to recognize the association of allergic phenomena with angina and myocardial infarction and be able to use a simple and inexpensive histologic technique to assist in postmortem diagnosis of Kounis syndrome.

This platform presentation will impact the forensic science community by increasing awareness of Kounis syndrome and providing information about a simple and inexpensive histochemical staining technique for use in the diagnosis of Kounis syndrome.

The association between acute coronary events and acute allergic reactions has been recognized for several years. The syndromes of allergic angina and allergic myocardial infarction, currently known as Kounis syndrome, is a rare condition first described in 1991. Recently, this syndrome has been linked to a wide variety of medical conditions including environmental exposures such as envenomations, medication exposures such as antibiotic administration in allergic patients, drug-eluted stent thrombosis, and coronary allograft vasculopathy. The presence of degranulated mast cells in the cardiac tissue along with elevated serum levels of histamine and tryptase is characteristic for this syndrome.

This study presents a case of a 49-year-old man with hypertensive cardiovascular disease who died immediately after being bitten by a copperhead snake (*Agkistrodon contortrix*). Histological sections of the myocardium revealed the presence of large numbers of degranulating mast cells in addition to the histopathologic changes characteristic of hypertensive cardiovascular disease. The presence of degranulating mast cells was confirmed by toluidine blue staining. The temporal relationship of the snake bite and sudden death of the decedent in addition to the histopathologic findings of degranulating mast cells in the myocardium are highly suggestive of a diagnosis of Kounis syndrome.

A study is being conducted using toluidine blue staining of heart sections of age-matched controls without a history of antecedent environmental exposure or evidence of an immune mediated response to assess the utility of toluidine blue staining as an adjunct in the histologic detection and quantification of degranulating mast cells in the myocardium and the specificity of those findings in allergic angina and allergic myocardial infarction.

This research suggests using toluidine blue stain as a readily available and inexpensive adjunct in the diagnosis of allergic angina and allergic myocardial infarction in decedents.

Kounis Syndrome, Snakebite, Mast cells

G118 Postmortem Examination of a High-Altitude, Diving-Related Fatality 17 Years After the Incident

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After attending this presentation, attendees will be familiar with the key elements of postmortem examination of diving-related deaths, understand the basic physics and physiology of SCUBA diving with an emphasis on the effects of altitude, and appreciate the importance of interpreting postmortem findings in the context of the events prior to death. They will also see a unique case of adipocere formation with remarkable preservation of the body and internal organs 17 years postmortem.

This presentation will impact the forensic science community by providing an example of a diving-related fatality due to an error in judgment in the setting of altitude diving.

Diving-related fatalities are rare occurrences. When they do occur, it is important for the forensic pathologist performing the postmortem examination to have an understanding of the physics and physiology of SCUBA diving and apply it to the circumstances surrounding the incident in order to determine the cause of death.

A body was discovered in Lake Tahoe, an alpine lake at a surface elevation of 6,225 feet in the Sierra Nevada mountain range. The body was found by recreational divers at a depth of 250 feet and brought to the surface by a robotic submarine. At the time of recovery, the body was clad in full dive gear, including a wetsuit with hood, gloves, booties, a buoyancy compensator vest with attached SCUBA tank and regulator, a 27-pound weight belt, and fins. The body was determined to be that of a 44-year-old male who had gone missing while diving with a friend 17 years earlier.

Postmortem examination revealed a partially skeletonized male with preservation of the torso and lower extremities by adipocere formation. No external evidence of antemortem trauma was identified. The internal organs were fairly well preserved due to encasement by adipocere, allowing examination of the cardiovascular system, lungs, liver, and kidneys. Moderate stenosis of the proximal left anterior descending artery was noted as well as mild calcific atherosclerosis of the abdominal aorta. Both lungs exhibited anthracotic pigmentation on the pleural surfaces.

Toxicology was performed on cardiac blood. Nicotine was detected, but there were no other positive findings of toxicological significance.

The equipment was sent to a dive expert and determined to be intact with the valve in the open position. Evaluation of the air mixture could not be assessed due to water in the tank, which appeared to have siphoned into the tank during his free-fall descent.

Review of the dive profile demonstrated that the divers were unfamiliar with altitude diving procedures. They dove to an actual depth of 100 feet with plans to stay at depth for a maximum of ten minutes. Approximately five minutes after arriving at depth, the friend noticed that his air supply was low, and he signaled to ascend. At a depth of 45 feet, he observed the decedent 25-30 feet below in free fall. He went after the decedent but was unable to remove the decedent's weight belt or put his mouthpiece into the decedent's mouth. At 130 feet, his air supply was significantly limited, so he returned to the surface alone. The buddy's equipment was examined at the time of the incident. Per the report, the

equipment was functioning normally, but the 72-cubic-inch tank was empty.

After thoroughly reviewing the postmortem examination findings, toxicology, and dive profile, the cause of death was determined to be drowning due to cardiac arrest from lack of air while SCUBA diving with coronary artery atherosclerosis listed as a contributing factor.

This case exemplifies the need for the forensic pathologist performing a postmortem examination in a diving-related fatality to thoroughly review the dive profile, to be knowledgeable about the physics and physiology of SCUBA diving, and to seek expert consultation when needed in order to ensure that a correct cause of death is provided.

Diving, Fatality, Adipocere

G119 Complete Forensic Investigation Reveals SUDEP and Not Homicide

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The goal of this presentation is to focus on a forensic case where an Italian public prosecutor initially suspected a homicide-related death. This presentation will examine a "Sudden Unexpected Death in Epilepsy" (SUDEP) event that required a complete forensic approach, i.e., clinical history collection, autopsy, and histological and toxicological studies.

The presentation will impact the forensic science community by demonstrating the significance of a thorough forensic investigation in reaching a correct postmortem diagnosis and, consequently, in providing an adequate and high-quality service to the judicial authorities. This presentation will also impact society as a whole by showing the crucial importance of protecting epilepsy sufferers, especially if they live alone. Particularly, an evaluation of the environmental conditions as well as periodic medical controls are recommended.

Epilepsy is a common chronic neurological disorder characterized by seizures and significantly increased rates of mortality. However, SUDEP, which is the most common seizure-related category of death, is still a poorly understood phenomenon. The precise frequency of its occurrence is not well defined so far, but a range of 1 in 370-1,100 in the general epileptic population is reported in the literature reviewed. From a pathophysiological point of view, it is possible to talk about SUDEP only in the absence of any identifiable cause of death at postmortem examination, suggesting an underlying arrhythmogenic predisposition or central/obstructive apnea or neurogenic pulmonary edema. Some authors have also suggested the key role played by inadequate therapy.

A 51-year-old Italian man was found lifeless, injured, and partially carbonized at his home during the winter of 2013. The man was supine, with his head inside the fireplace (the fire had recently gone out) with multiple and diffuse second-, third-, and fourth-degree burns mainly placed in the posterior regions of the body. The victim had the typical "pugilistic position" with areas of split skin mixed with burn lesions. The occipital area showed an irregular cranial discontinuity (diameter: 1cm; depth: 1.5cm) without surrounding skull fractures. A right collar-bone fracture was also observed. During the crime scene investigation, the house was

in considerable disorder and the victim's brother-in-law, who was questioned by police as an informed witness, presented, according to police officer reports, with "suspect injuries." The combination of these data led the public prosecutor to hypothesize that the man might have been killed during an intra-familial robbery attempt.

A forensic autopsy was performed within 48 hours in order not only to find the real cause of death, but also to investigate the hypothesis of homicide. The internal examination mainly revealed: "heat-hematoma," no discontinuity of the dura mater, boiled cranial content, absence of internal injuries and/or hemorrhagic infiltrations, no smoke in the upper airways, and pulmonary edema. The evaluation of histological specimens obtained from autopsy samples showed massive edema of the lungs associated with multi-organ congestion, liver steatosis, and no vital signs in the epidermal lesions. Toxicological analysis resulted minimally positive for carboxyhemoglobin (COHb) (8.5%) with a complete absence of alcohol, phenobarbital, and illegal drugs in blood and urine.

Moreover, the forensic pathologist, on analyzing the victim's medical history, discovered that he had suffered from epilepsy (treated with phenobarbital, though with poor medication compliance) with recurrent falls (some of them leading to traumatic injuries clearly shown in previous X-rays and Computed Tomography (CT) images; a therapeutic craniotomy was also performed in 2006). The brother-in-law also underwent a forensic examination which resulted positive due solely to recent tattoos.

Only by performing such a complete forensic investigation was it possible to exclude with certainty the initial suspicion of homicide and to make a final diagnosis of SUDEP. In fact, the victim was undoubtedly subjected to the action of the flames when he was already dead or *in limine vitae*.

It could therefore be concluded that the victim fell into the fireplace after the verification of SUDEP, resulting in postmortem burn injuries. The occipital discontinuity was also explained by the deceased's prior medical history (craniotomy).

This case underlines the need to keep attention levels high in the forensic analysis of all so-called "seizure-related deaths," such as burns, accident, suicide, treatment-related deaths, and SUDEP.

SUDEP, Forensic Investigation, Differential Diagnosis

G120 Family Mass Murder: A Cluster of Four Cases in Apulia, Italy, and a Review of the Literature

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After attending this presentation, attendees will understand the main characteristics of familicide, a rare and poorly understood form of homicide.

This presentation will impact the forensic science community as it shows that family mass murders represent an important part of domestic homicides, especially in Italy where the value of "family" has declined.

Family mass murder or familicide is a subtype of domestic homicide. The latter represents murders in which the perpetrators and the victims are both men and women with different degrees of kinship. The former is an enlarged form of intrafamilial homicide with multiple killings, usually involving the offender's spouse, one

or more of the children, and, sometimes, the offender himself. Few documents provide an in-depth study of familicides because public interest and crime statistics tend to focus more on domestic/ intrafamilial homicides in general than on specific and rare forms such as family massacres.¹ In Italy, the Home Office (Minister of Interior) and the security forces do not have a national registry for familicide incidents, and neither does the National Institute of Statistics (ISTAT). What is known is that 155 incidents of domestic homicide occurred in Italy in 2012 (72 intrafamilial), 113 in 2010 (81 intrafamilial), 122 in 2009 (97 intrafamilial), and that they took place more often in Northern Italy.² Between 1985 and 2008, 89.5% of homicide-suicide mass murders in Italy involved family situations, and in all cases of familicide between 1990 and 2009 in the province of Milan, firearms were used.^{3,4}

Familicide may be perpetrated by any family member, but most often involves the male head of the household. It is often carefully planned in advance and very often followed by suicide. The event is usually preceded by a gradual but steady increase in stress due to financial or personal factors and is precipitated by an event such as loss of a job or divorce that represents the final, unbearable stressor. Occasionally, the pets are also killed and the house is set on fire.

This presentation describes four cases of familicide-suicide in which the perpetrator was the husband and father of the victims and no apparent stressor had impacted the men or their family's lives.

The first case took place in March 1990 in Bisceglie. The husband was a 60-year-old man who had recently moved back to Italy with his family. The man stabbed his 66-year-old wife in the abdomen and in the back and then stabbed and shot the 24-year-old daughter as well as the family dog which was found close to the girl. He barricaded himself in the house and the next day he committed suicide by cutting his left elbow and forearm and shooting himself in the head.

In March 1991, the second familicide happened in Bari. A 32-year-old man killed his 35-year-old wife and his children (4- and 8-years-old). He stabbed them in the neck, but the children did not die after this initial assault. He ultimately finished the killing by manual strangulation. He was convicted of the crime and committed by hanging suicide one week later in jail.

The third case dates back to March 2008 in Taranto, when a 48-year-old vascular surgeon killed his two daughters (11- and 14-years-old) with hammer blows to the head while his 43-year-old wife, tied to the bed, was listening to the screams of her children. He then reached the wife and struck her on the head with the same hammer used on the daughters. After the massacre, he took his own life by cutting his femoral artery with a scalpel.

The fourth case occurred in May 2013 in Sannicandro. A 54-year-old pharmacist shot his 58-year-old wife and 19-year-old daughter (who had Down's Syndrome) in the head while they were sleeping in their beds. He then waited in the living-room for his 24-year-old son to come home from his job. When he entered the house, the father shot him in the head from behind. After the massacre, he hid the gun and drowned himself in the swimming pool. When police arrived, only the son was still alive, but he died shortly thereafter in the hospital.

This presentation will analyze each case in detail and review literature about familicide.

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Familicide, Homicide/Suicide, Mass Murder

G121 Death of an American Abroad—Forensic Opinion Hanging by a Thread

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After attending this presentation, attendees will: (1) appreciate that in a globalized world, death investigation is no longer a local jurisdictional concern. Death of foreign persons can sometimes take on larger dimensions, even on an apparently straightforward case; and, (2) recognize that the ability to conduct an autopsy does not a forensic pathologist make. One who claims to provide a second forensic opinion must carry out due diligence in the process by examining all the evidence available, distinguish what is fact, opinion, and speculation, as well as recognize the limit of his or her expertise.

This presentation will impact the forensic science community by highlighting the implications of unqualified persons expounding forensic opinion in the practice of forensic pathology, and the damage such opinions can cause.

An American scientist with a PhD degree was employed by a government-related research agency in Singapore. At the end of his employment contract, just prior to his departure to go back to the U.S., he was discovered hanging in his residence.

Even as routine investigations into the circumstances of his death were underway, several allegations were made and published in a global mainstream newspaper article, alluding to possible homicide by a foreign spy agency. These allegations were backed up by a expert's forensic opinion, disputing the initial cause of death and calling the cause of death garroting, and manner of death homicide. The case was subsequently picked up by other media outlets and eventually led to the involvement of the Federal Bureau of Investigation (FBI), politicians, and high-level government officials from both the U.S. and Singapore.

The coroner's inquiry was conducted in public, with five legal counsels acting for the next-of-kin, and attendance of a number of U.S. media representatives in court. A total of more than 70 witnesses were called, including the consultant pathologist who supervised the case, the family's expert, and two other independent forensic pathologists.

The presentation will cover the facts of the case, focusing in particular on the autopsy and forensic findings which formed the core of the dispute between the experts.

The presentation will highlight the fact that forensic death investigation is no longer purely a local jurisdictional matter, that in an age of globalization, internet, and social media, matters can get out of hand very quickly. Deaths of foreigners can have an added dimension that may not usually be associated with "local" cases. Secondly, the case will demonstrate that unqualified forensic opinion can turn an apparently straightforward case into a media spin.

Hanging, Strangulation, Expert Opinion

G122 Information System of the Allegheny County Medical Examiners' System

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The goal of this presentation is to illustrate to the attendees the need and utility of a modernized, electronic information management system in a medical examiner's office.

This presentation will impact the forensic science community by demonstrating how a modern electronic information management system can reduce error and improve communication in the office and between related agencies outside the office.

Background: The medical examiner's office is a complete death-investigation facility, including a fully equipped autopsy suite and state-of-the-art forensic laboratories. The complete operation consists of many investigation and autopsy personnel, where forensic investigators investigate death calls and retrieve bodies from death cases, and forensic pathologists and autopsy technicians perform postmortem examinations. The complete forensic laboratory consists of forensic biology, drug chemistry, toxicology, firearms and tool marks, trace evidence, latent fingerprints, the mobile crime unit, and the mobile laboratory. These laboratories utilize the expertise of dozens of additional specialists. In recent years, the increased complexity and sophistication of the system necessitated a more streamlined system of communication and data sharing between the various branches and personnel. Part of this initiative involved implementing an electronic information management system which, at a minimum, needed to be able to assign case numbers, track the evidence associated with the case, and provide a printable chain of custody for the evidence. Ultimately, this information system has evolved into a more comprehensive system of data management and communication.

Methods: Over the course of three to four years, the medical examiner's office implemented a Porter Lee's Barcode, Evidence, Analysis, Statistics, and Tracking (BEAST) information management system. The BEAST has two components: a Laboratory Information Management System (LIMS) and the Medical Examiner Information Management System (MEIMS). The BEAST is used to maintain basic case information, track the location and custody of items recorded into the system, record results of laboratory analysis and death investigations, store external/outside documentation related to the case (medical records, third-party test results, etc), create notes, reports, and forms for each case, generate statistics, track courtroom activity, interface with instruments to export information to the instrument and import results from the instruments, and host links to other laboratory information areas, such as Photov Vault® and SharePoint®. Photo Vault® is used to store scene images and autopsy photos. Outside investigating agencies have limited access to the Photo Vault® to view scene and autopsy photos. SharePoint® is used to store the policy and procedure manuals for the entire office. It is separated into sections to allow each department to have their own workspace, calendar, document storage, and reference storage.

The BEAST also has a web portal hosted by Pennsylvania Justice Network (JNET) that allows police agencies to enter evidence they wish to submit to the laboratory, view the progress of their case through the laboratory, and download approved copies of laboratory reports. The district attorney's office has been granted access to the portal to allow for the download of reports. This is

done from a computer that has JNET access and a user that has an authorized user ID and password.

Results: Pathologists now have the capability of accessing all relevant forensic data regarding each case from their office workstation. This has resulted in time-reduction of case signout as well as more comprehensive reviews of complex cases without the need to communicate personally with other divisions. Furthermore, personnel from all medical examiner office divisions, as well as those from some outside agencies, now have direct access to needed information without significant loss of time.

Conclusions: The implementation of this information management system has proved beneficial in many respects. The medical examiner's office has improved the tracking of files and evidence, increasing the productive efficiency and allowing rapid and efficient sharing of information within the office and with outside agencies as well.

Information System, SharePoint®, Beast

G123 The Technical Working Group Postmortem Angiography Methods (TWGPAM): An International Collaboration for Research and Training

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After attending this presentation, attendees will be familiar with the the Technical Working Group Postmortem Angiography Methods (TWGPAM), an international working group created in February 2012 in Lausanne, Switzerland. Attendees will know its structure, the aims of the group, and its ongoing projects.

This presentation will impact the forensic science community by providing information about the TWGPAM, a recently developed research group that is very active in performing multi-center research, organizing workshops, and providing mutual assistance for centers starting to implement CT-angiography. This presentation gives an overview allowing attendees to obtain information about the possibilities offered by the group.

Postmortem Computed Tomography- (CT) angiography (pmCTA) is an exam that seeks to increase the sensitivity of postmortem investigations by providing useful information about the vascular system and its lesions. Although research conducted currently proves that this goal can be achieved, most applied methods have remained experimental for the moment, and centers using pmCTA as a routine exam are few. In order to become a technique that is accepted in the forensic scientific community and in jurisprudence as a proof in court, standardized and validated methods are necessary. To optimize the workflow and integrate the exam into daily routine in a medicolegal center, adequate equipment and formation of staff is necessary. To satisfy these needs and to further develop and validate techniques of pmCTA, an international working group called TWGPAM was created in 2012. Today, it consists of nine participating centers situated in six European countries: Switzerland (University of Lausanne and University of Basel); Germany (University of Hamburg, University of Munich and University of Leipzig); England (University of Leicester); Italy (University of Foggia); Poland (University of Krakow); and France (University of Toulouse). Each center provides a team consisting of forensic pathologists, radiologists, and radiographers. The teams meet regularly three times a year for meetings to exchange experiences and discuss ongoing studies and future strategies.

Between these meetings, the different teams exchange data with each other and report cases on a secured homepage, generating a growing common database and allowing a constant exchange of the increasing knowledge between members.

As a first research goal, the TWGPAM group has started a multi-center study in order to validate the technique of Multi-phase Postmortem CT-Angiography (MPMCTA). In the context of this study, each center expected to investigate about 50 cases using the standardized protocol of this method and standardized equipment. The idea of this study is to definitely identify advantages and limitations of the method and to define indications for its application. Once this first multi-center study is finished, further studies are planned allowing the validation of other techniques as well as the development of new protocols for performing pmCTA.

Besides performing different studies, one primary goal of the TWGPAM is the dispersion of knowledge. Therefore, the TWGPAM members are organizing workshops at international meetings and a pmCTA workshop will be held once a year. The next challenge for the team is the editing of an "Atlas for Postmortem Angiography" which should help physicians all over the world start their own pmCTA exams and will provide guidelines for the interpretation of radiological images.

Postmortem CT, Postmortem Angiography, Forensic Imaging

G124 Investigating the Order of the Dual Examination of Touch Prints Via Fingerprint and DNA Analyses

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After attending this presentation, attendees will gain an understanding of the effects that a dual examination of a single piece of evidence for latent prints and DNA have on one another.

This presentation will impact the forensic science community by providing information on the effects of collecting latent prints and DNA from a single piece of evidence.

Latent print development for investigational leads has been established in the criminal justice community for a long time. However, with recent advancements in DNA profiling, obtaining DNA samples from latent prints has become a reality. These two processes together can provide valuable information to a forensic investigation. However, determining the order in which the analyses are done has not been thoroughly investigated. A comparison of samples analyzed first for DNA and second for fingerprints versus an analyses conducted in the opposite order can provide insights into which technique should be given priority when examining latent prints.

A standardized sample of 100 fingerprints and low-template DNA from a single individual was prepared by pipetting 20uL of saliva onto the designated fingerprint area on plain, white computer paper. Once dry, a right thumbprint was made by using an amino acid-based latent print reference pad. Half of these fingerprints were subjected to DNA extraction first, followed by latent print processing. The other half were processed for prints and then swabbed for DNA using the double-swab method. A Millipore® Amicon extraction protocol was used to extract the DNA and this DNA quantified using the Quantifiler® kit. The fingerprint samples were then analyzed by a fingerprint analyst at the Indiana State Police to determine their usability in an investigation. The analyst used both 1,8-Diazafuoren-9-one (DFO) and ninhydrin in the examination which is standard protocol for fingerprint analysis at the Indiana State Police. The resulting prints were then evaluated

to determine the quality of the sample.

The average quantity of DNA when DNA was first extracted was $1.126 \pm 0.789\text{ng/uL}$ and $0.932 \pm 0.866\text{ng/uL}$ when the DNA was extracted following latent print processing. The fingerprint analyst determined that of the 50 samples examined, nearly half of them were suitable for use in fingerprint examination. For the remaining 50 fingerprints, all 50 were considered usable for fingerprint analysis.

The results obtained from this investigation can be integral in increasing the efficiency of overall use of latent prints. By determining the best order for completing a dual exam, forensic scientists can effectively acquire results related both to fingerprint and DNA analysis. The results listed above indicate that the amount of DNA collected during each of these trials did not differ significantly. With the information obtained in this research, the forensic science community can more effectively collect information related to latent prints and DNA from a single piece of evidence.

Latent Prints, DNA Extraction, DNA Quantitation

G125 Status of National Policy Initiatives for Forensic Science Reform

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After attending this presentation, attendees will: (1) know the various policy options being discussed and their differences; (2) understand the implications of each policy; and, (3) critically consider each policy option being discussed.

This presentation will impact the forensic science community by increasing awareness of the fact that the result of the current policy efforts will likely result in federalization, uniformity, raising standards, increased compliance requirements, and unfunded mandates.

Since the 2009 National Academy of Sciences (NAS) Report, *Strengthening Forensic Science in the United States: A Path Forward*, there have been a number of policy initiatives set in motion such as the White House Subcommittee on Forensic Science, the Leahy Senate Bill introduced, the Department of Justice/National Institute of Standards and Technology (DOJ/NIST) National Commission on Forensic Science announced, and the Scientific Working Groups (SWGs) set to transition.¹ Although there are significant hurdles that may prevent their coming to fruition, it appears likely that these efforts will result in a policy shift. Law enforcement and public safety, including crime laboratories and medical examiner and coroner offices, has traditionally been the province of the state and local governments. However, this has resulted in a patchwork of systems with varying quality. Although there is value in experimentation among the pluralist state systems and there is value in tailoring systems to local needs and local communities, these justifications for the current local control seem to be trumped by the argument that a perpetrator should not get away with murder by crossing a jurisdictional boundary. Critics of the current system, defense attorneys, and many forensic scientists have called for national standards. Although professional organizations in some disciplines (National Organization of Medical Examiners (NAME), for example) have developed national standards, others have not. Given the lack of standards in some disciplines, the Federal Bureau of Investigation (FBI) and the National Institute of Justice (NIJ) have historically sponsored SWGs. However, the NAS Report criticized the SWGs for their haphazard quality and other shortcomings. It is not clear if the current federal government-sponsored SWGs are governmental, quasi-governmental, or non-governmental entities, but it is clear that they

are not fully compliant with the Office of Management and Budget (OMB) Circular A-119 on federal participation in the development and use of voluntary consensus standards and conformity assessment activities nor are they currently able to enforce any standards that they might promulgate. The number of SWGs has swelled to 21, to include disciplines that have professional organizations which have promulgated standards, and there has been an effort to standardize the SWGs by a SWG chair's meetings. It appears that the SWG administration will transfer from DOJ to NIST. The American Society for Testing and Materials (ASTM) E-30 committee has been promulgating standards in compliance with OMB Circular A-119 with mixed community acceptance. The DOJ/NIST National Commission on Forensic Science would use the SWGs or replace them with discipline-specific "guidance committees" whose reports may be considered by a National Commission that reports to the Attorney General, who would have ultimate authority over any policy recommendations. The White House Subcommittee on Forensic Science appears to have fed into the development of the National Commission which would be composed of many different stakeholder groups. The Leahy bill, on the other hand, would replace the SWGs with discipline-specific "committees" composed entirely of scientists, which would make recommendations to a Forensic Science Board. In this latter case, the Board would not be able to amend the recommended standards without re-consulting the committee. Also, under Leahy, the DOJ could decide against any recommendation of the Board, but this would trigger review by Congress. Congress does not seem to have the appetite to fund committees that are as large as the current SWGs, nor an overly large supporting bureaucracy. Both the White House-derived National Commission and the Leahy Forensic Science Board would have setting standards as a primary mission. Unlike the National Commission, the Forensic Science Board, which would be backed by legislative authority, would have the "rule-making" power to impose its standards on state and local governments. The Leahy bill would mandate accreditation and certification; this is probably a goal of the National Commission as well. The Leahy bill also seeks to create a national ethics code, standardization of terms, and a research program. Cost and/or politics could thwart these initiatives. If implemented, these initiatives would result in some degree of regulation and federalization of the forensic sciences. Essentially, these efforts would be a federal unfunded mandate that the state and local forensic science agencies bring their performance up to a national set of standards. It can be envisioned that some agencies and some managers will be unable to comply while others will prosper.

Reference:

1. National Academy of Sciences, *Strengthening Forensic Science in the United States: A Path Forward*, National Academies Press, 2009.

Policy, Regulation, Federalization

G126 Scientific Working Group on Disaster Victim Identification (SWG DVI) Update

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After attending this presentation, attendees will: (1) recognize efforts being taken to establish guidance for mass fatality operations; and, (2) understand concepts of fatality management that undergird developing standards.

This presentation will impact the forensic science community by informing local medical examiners and coroners to be aware that guidelines for fatality management are being

developed.

The Scientific Working Group on Disaster Victim Identification (SWG DVI) was formed to advance the scientific basis for disaster victim identification by assembling professionals from the DVI community, including international participants, in a collaborative effort to exchange ideas regarding scientific analysis methods, protocols, training, and research related to DVI. The first meeting of the SWG DVI convened in the summer of 2010. The Committee has grown to 31 members and has retained international membership. Permanent members of the Board include National Association of Medical Examiners (NAME), the International Association of Coroners & Medical Examiners (IAC&ME), the National Transportation Safety Board (NTSB), the Federal Bureau of Investigation (FBI), the Department of Health and Human Services Assistant Secretary for Preparedness and Response (DHHS (ASPR)), the Department of Defense Armed Forces Medical Examiner System (DOD (AFMES)), the International Criminal Police Organization (INTERPOL), the International Committee of the Red Cross (ICRC), and the International Commission on Missing Persons (ICMP) and is supplemented by individual members with varying specific expertise and experience. The Board oversees 11 committees: Search and Recovery; Odontology; Molecular Biology/DNA; Friction Ridge Analysis; Pathology; Anthropology; Data Management; Victim Identification Center/Family Assistance Center; DVI Reconciliation and Quality Assurance; DVI Management; and, Ethics. The SWG DVI Board recommends that jurisdictions predetermine a set trigger point, e.g., suspicion of ten victims — initiate DVI operations. The SWG DVI Board believes that successful disaster victim identification efforts begin with, and are reliant upon, efficient, effective, and timely search-and-recovery operations. Documentation of the place and position of the remains may be important for identification and investigation. Hence, the Board believes that those medicolegal authorities that rely on others to recover remains, particularly without their own office input or involvement, are doing their jurisdiction a disservice. Data collection centers, such as call centers, are important to set up and assist with the development of good victim manifests. The Victim Identification Center (VIC) is a component of the Family Assistance Center (FAC) in which families and loved ones are gathered to obtain antemortem data, including DNA specimens, to compare against the postmortem data developed in the morgue. Postmortem data is obtained from pathologists, anthropologists, friction ridge analysis, and molecular biology/DNA testing forensic science experts. Data management systems are crucial to large-scale operations to organize and utilize the data. The American National Standards Institute-National Institute of Standards and Technology (ANSI-NIST) (500-290) format standards and the National Information Exchange Model (NIEM) semantic standards have been developed which may permit systems to exchange DVI biometric data. Outside database searches are also important; fingerprints, dental, and DNA data may be searched against several databases. A common mistake is to rely solely on a single fingerprint database search. Eventually, the data must be reconciled to develop the identification for every human remain. The entire set of DVI operations must be managed, financially documented, and quality assured. Families must be kept informed of the progress of the DVI operations. The ethical principles that must be considered during all DVI operations are individual autonomy, human dignity, and respect for persons. After three years in development, specific guidelines and best practices are being promulgated by the SWG. These documents, available on the SWG DVI website, are intended to be of use to all medicolegal authorities and those who interact with them. The SWG DVI hopes that these documents will be consulted and be of value during the development of mass disaster plans as well as serve as references for fatality management operations when an incident occurs.

G127 Medical Examiner and Forensic Lab Operations During a Natural Disaster: Lessons Learned From Hurricane Sandy

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After attending this presentation, attendees will learn more about Disaster Victim Identification (DVI) and some of the challenges faced by the New York City Office of Chief Medical Examiner (NYC OCME) during Hurricane Sandy and its immediate aftermath.

This presentation will impact the forensic science community by reviewing the plan used by the medical examiner's office to identify victims of Hurricane Sandy as well as maintaining continuity of operations immediately after the storm. The community will also hear about lessons learned.

On October 29, 2012, a Category 1 hurricane made landfall near Brigantine, New Jersey. The storm surge hit New York City, flooding streets, tunnels, and subway lines. Power was cut in and around New York City leaving approximately 1.5 million people in New York in the dark. Schools were closed for a week, cell phone service in many parts of New York City was non-operational, traffic signals were not working on many New York City streets, bridges were closed, mass transit and airports were shut down, and fuel shortages were commonplace. City shelters were opened throughout New York City for displaced persons, the New York Stock Exchange was closed for two consecutive days, and two area hospitals had to be evacuated. There were 44 direct storm-related fatalities and approximately \$20 billion in property damage in New York City.

The NYC OCME activated its All-Hazard Mass Fatality Management Plan and transferred all of its regular daily death cases to its Queens facility. Disaster morgue operations were located at the OCME's Manhattan facility and due to the rapid rate of recovery, operations were able to be run close to normal. NYC OCME field operations were set up in the Rockaways, Queens, and Staten Island. Most fatalities were located in low-lying coastal areas of Staten Island and the Rockaways with the cause of death being 79% drowning and 14% blunt impact trauma. Ninety-three percent of victims were identified through contextual means and 5% were identified using fingerprints. No DNA or dental records were required to make identifications. NYC OCME was successful in managing simultaneous continuity of operations and external disaster responses.

This presentation will also review NYC OCME's Continuity of Operations Plan (COOP) and the importance of identifying other skill sets that employees may have prior to any disaster. In the aftermath of the storm, NYC OCME employees were instrumental in a variety of diverse tasks in logistics and operations.

The NYC OCME experienced some significant equipment and facility damage. Moreover, in a major catastrophic event, city agencies may need to be totally self-sufficient. Lessons learned by the NYC OCME in the aftermath of Hurricane Sandy can help other medical examiner offices and crime laboratories better develop their own disaster management plan.

Hurricane Sandy, Disaster Victim Identification, Natural Disaster

G128 Death From WD-40™ Ingestion

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After attending this presentation, attendees will understand the pathophysiology of hydrocarbon ingestion deaths and the importance of multidisciplinary consultations in casework.

This presentation will impact the forensic science community by familiarizing attendees with the pathophysiology of deaths resulting from hydrocarbon ingestion and the importance of multidisciplinary consultations in casework.

Hydrocarbon ingestion fatalities are uncommon and case investigations benefit from the effective synchronization of efforts, including active cases. Although hydrocarbon exposures are not uncommon (>43,000 cases in 2009, with 60% in teens and younger), deaths are rare (9 or ~0.02%) and usually associated with the very young (<5 years old) and adolescent abusers. Exposures typically result from inadvertent ingestion by children or complications stemming from inhalational abuse. Common ingestions include gasoline, kerosene, lubricating oil, motor oil, and many others, of which WD-40™ (a lubricant, penetrating oil, and water-displacing) is one. The active ingredient is a non-volatile, low-viscosity hydrocarbon whose primary ingredient is listed as Stoddard solvent; however, the exact formulation is secret. Recognized potential hazards include irritation (skin, eyes, and especially respiratory), narcosis, and renal impairment.

Case Study: At ~3:00 a.m., a 57-year-old, 74½-inch, 287.4-pound male was found by his wife at his residence, retching. He returned to bed but at ~6:30 a.m. complained of respiratory difficulty and was transferred to the local hospital where his condition deteriorated and he was pronounced dead at 6:30 p.m. Hospital staff became concerned because the patient's emesis and skin became green. Eventually, the wife revealed the the decedent might have ingested WD40™ that he kept at the house. At autopsy, the tissues and gastroenteric contents had a mild non-specific chemical odor. The stomach and bowels contained a clear, mildly pale-green fluid. The bowels had apparent sloughed mucosal surface. The tracheobronchial tree contained dull gray-green mucoid material. The hepatic parenchyma had gross necrosis and the kidneys were variegated. Multiple variably prominent petechial hemorrhages were present. Other significant findings at autopsy included concentric left ventricular myocardial hypertrophy (580 grams). The cause and manner of death were certified as suicidal petroleum distillate ingestion — without toxicology results.

Subsequent analysis for volatile organic compounds by headspace gas chromatography revealed.*

* Other than the described findings, examination of the specimens submitted did not reveal any positive findings of toxicological significance

**A pattern of aliphatic and aromatic hydrocarbons consistent with the fluid sample submitted was detected. In addition, a second group of aliphatic and aromatic hydrocarbons with less volatility was observed in this specimen that was not detected in the fluid sample.

Source	Hydrocarbons (fuel oils)	Hydrocarbons (compared with submitted fluid)	Ethanol (mg/dL) (mg/100 gm)	Acetaldehyde (mg/dL) (mg/100 gm)	Acetone (mg/dL) (mg/100 gm)
Femoral blood (autopsy)	C21 through C27	N/D	64	4.4	Trace
Gastric content	C12 through C15	Positive**	29	1.0	1.3
Small bowel content	C12 through C15		N/D	N/D	N/D
Large bowel content	C12 through C15		N/D	N/D	N/D
Liver	N/D	N/D	24	36	N/D
Lung	N/D	N/D	N/D	N/D	1.2
Unknown fluid	C9 through C11	Positive			
WD-40™ (purchased)	C9 through C11				

Hydrocarbons are aliphatic (non-ring) or aromatic (ring) with toxicity directly related to the agent's physical properties and the extent dose and route dependent. The most common adverse consequence is aspiration injury, with lower viscosity hydrocarbons associated with increased risk. Aliphatic hydrocarbons have little gastrointestinal absorption and commonly result in emesis with aspiration and a secondary severe chemical pneumonitis. Aliphatic hydrocarbons easily cross the blood-brain barrier and can have acute or chronic effects, depending on the nature of the exposure. Sudden death may be associated with cardiac tachydysrhythmias. One-third of cases are associated with emesis; hydrocarbons are a direct mucosal irritant. The liver may have centrilobular zonal necrosis and lipid peroxidation. Certain chronic renal exposures may have anion gap acidosis. Hematologic abnormalities may include hemolysis (acute) and aplastic anemia, multiple myeloma, and acute myelogenous leukemia (chronic).

In this case study, death was associated with hepatic necrosis and chemical pneumonitis as delayed subacute features of a suicidal ingestion. The communication between the toxicologists and pathologist allowed fairly rapid completion of the death certification process based on scene and historical information without toxicology results, which were delayed for an extended period of time.

Hydrocarbon, Toxicology, Pathophysiology

G129 Sudden Death Following Mono-Intoxication With 3,4-Methylenedioxypropylvalerone (MDPV)

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The goals of this presentation are to understand the mechanisms of death following 3,4-Methylenedioxypropylvalerone (MDPV) use, the toxicology of MDPV use, and the pathology associated with MDPV use.

This presentation will impact the forensic science community by detailing the pathology and toxicology associated with an emerging drug.

"Bath salts" are the name given to a group of drugs that have been growing in popularity as recreational drugs in a number of regions in the world in the last few years. "Bath Salts" are synthetic cathinones and commonly encountered drugs include mephedrone, methylone, and 3,4-methylenedioxypropylvalerone (MDPV). Most reports have involved the drug mephedrone. Synthetic cathinones have been associated with multiple effects including panic attacks, anxiety, paranoia, psychotic behavior, hallucinations, aggressive behavior and excited delirium, insomnia, depression, and suicide. Reported physical effects include tachycardia, hypertension, sweating, hyperthermia, myocardial infarction, sweating, rhabdomyolysis, and seizures. Deaths have occurred.

A 42-year-old man had a history of a psychotic mood disorder and was prescribed olanzapine, citalopram, and clonazepam, but was thought to have run out four days before his death. He was found in the afternoon lying on the floor and talking incoherently. He was brought to the ambulance walking with assistance, but suffered a cardiac arrest in the ambulance. He was recorded in the ambulance as having a core temperature of 41°C (105.8°F). The autopsy revealed no significant injuries. On internal examination, there was non-specific congestion of the organs but no significant natural disease. Microscopic examination of the lungs revealed intra-alveolar and perivascular macrophages

with foreign birefringent material present. In the liver, there were increased neutrophils in the sinusoids, microvesicular fatty change, and pigment granules seen in macrophages. Toxicologic analysis revealed traces of MDPV along with traces of citalopram. Acetone was present in the urine. Subsequent quantitation revealed an MDPV concentration of 5.8mg/L.

The features in this case are that of hyperpyrexia following use of MDPV. No other drugs were present in significant amounts. The synthetic cathinones have been associated with the development of hyperpyrexia with core temperatures over 41°C (105.8°F). The administration of drugs via snorting (insufflation) is recognized, but pathological evidence of past use is rarely evident at autopsy. "Bath Salts" are reported to often be snorted.

Recreational drugs are often used in combination with other drugs. Reports of deaths with synthetic cathinones often involve multiple drugs with pharmacological activity. MDPV concentrations have been reported to be up to 8.0mg/L in drivers, illustrating again that recreational drug concentrations typically overlap with concentrations seen in fatal cases.

In conclusion, this case involves a mono-intoxication with MDPV causing fatal hyperpyrexia with evidence of past use of drugs by insufflation.

MDPV, Toxicology, Pathology

G130 Death Following Ingestion of Compounded Topical Cream

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The goal of this presentation is to describe a death and the associated autopsy findings following suspected ingestion of a compounded topical cream prescribed for the treatment of neuropathic pain.

This presentation will impact the forensic science community by describing how a seemingly safe topical pain medication could be lethally abused.

Topical application for the management of pain has become increasingly utilized because it has a major advantage of minimizing side effects due to lower systemic drug absorption. Additional advantages include direct delivery and increased drug concentrations to the pain source, reduction of possible drug interactions, ease of delivery, and better compliance for patients with difficulty swallowing pills. Topical creams come in various formulations and may be "compounded" or customized based on clinical symptoms. These may include combinations of opioids, tricyclic antidepressants, anticonvulsants, local anesthetics, and alpha-2 adrenoceptor agonists. When used appropriately and as prescribed, topical creams typically do not cause death. However, there is limited documented evidence on the lethality of these topical creams when ingested or delivered through a source other than the skin.

The following novel case study involves a 30-year-old male with a history of jaw pain secondary to an unresolved jaw fracture from remote trauma. His medical history included depression and anxiety. His social history was significant for polysubstance dependency and multiple opioid overdoses. He had been treated for his substance abuse in both inpatient and outpatient settings. His previous medications included methadone, oxycodone, gabapentin, fentanyl, various muscle relaxants, and anti-depressants. At the time of his death, he was being treated by a psychiatrist, an internist, and a pain specialist. Following a recent

meeting with his pain specialist, he was prescribed a topical cream containing ketamine (10%), cyclobenzaprine (4%), gabapentin (6%), tramadol (8%), clonidine (0.2%), and amitriptyline (4%). The cream was formulated by a compounding pharmacy and was dispensed in an airless metered dosing pump (40 grams total weight). It was sent to the decedent's residence and the decedent signed for the package himself. Two days later, the decedent was found unresponsive in his bed following multiple failed attempts to reach him. Scene investigation was significant for a postmortem rectal body temperature of 106°F. The metered dosing pump containing the cream was found at the scene and, based on the calculated weight of the container and the substance, deemed to be empty.

At autopsy, the decedent was of appropriate build and nourishment and external examination was unremarkable. Internal examination revealed approximately 300mL of gastric contents in the left pleural cavity. Further examination revealed defects of the stomach and diaphragm suggestive of gastromalacia. There was no gross color change of surrounding organs evident. The remainder of the examination was unremarkable. Histologic examination showed no inflammation or any vital reaction of the pleura. Toxicological examination revealed femoral blood concentrations (mg/L) as follows: amitriptyline 0.28; nortriptyline 0.47; tramadol 0.89; cyclobenzaprine 0.42; ketamine 0.15; and gabapentin 5.40. Oxycodone and oxymorphone were detected in the urine, but not the blood. Gastric contents were retained and re-examined at a later time. After thawing, a viscous, dark yellow and granular substance was present at the top of the tube. Toxicological examination on a portion of total gastric contents revealed concentrations (mg/L) as follows: amitriptyline 225; tramadol 214; cyclobenzaprine 217; ketamine 239; and gabapentin 132.

Although considered safe and unlikely to cause significant systemic concentrations of drug, medical examiner's must always consider the possibility that these topical creams are potentially ingested or delivered through another mucosal source and can lead to significant systemic absorption. This must be considered especially in circumstances where the recipient has a known history of substance abuse. Careful examination of gastric contents and other mucosal surfaces must be correlated with scene examination and clinical history.

Topical Pain Creams, Compounded Creams, Gastromalacia

G131 Pulmonary Hypertension in Users of Levamisole-Contaminated Cocaine

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After attending this presentation, attendees will learn how the use of levamisole-contaminated cocaine may lead to the occurrence of idiopathic pulmonary hypertension.

This presentation will impact the forensic science community by raising awareness of the potential lethal consequences that may result from chronic levamisole ingestion and make the community aware of a new public health threat.

Background: The conversion of levamisole to aminorex in horses was first observed in 2009. Two years later, there was laboratory confirmation that the same conversion occurs in humans. Since then, Maurer and his group in Germany have

isolated five different isomers of the two compounds. Both drugs interfere with serotonin metabolism. It has even been speculated that cocaine is being intentionally adulterated with levamisole to increase its psychogenic effect. Aminorex was once sold in Europe as an anorectic, but was withdrawn from the market after a cluster of deaths from Idiopathic Pulmonary Hypertension (IHP) were reported, mostly in women attending weight-loss clinics.

Objectives: Various monitoring agencies now report that most of the world's illicit cocaine supply is contaminated with levamisole. Some estimates suggest that as much as 70% of street cocaine may be contaminated. This raises the possibility that users of levamisole-adulterated cocaine may be at risk for IHP. This presentation describes the autopsy and toxicology findings of the first such case.

Materials and Methods: A complete autopsy was performed on an individual who died of heroin toxicity and was also a known cocaine user. Peripheral blood samples were submitted for toxicological analysis. In addition, samples of urine, hair, and blood were all screened for the presence of levamisole and aminorex.

Results: Microscopic examination disclosed the classic findings of IHP, though no evidence of thrombotic arteriopathy was evident, in spite of the heroin use. Interlobular septal veins had become muscularized, and there was marked capillary distention in periseptal alveoli. Some areas displayed marked vein wall thickening and decreased lumen. Adjacent alveoli were sometimes filled with blood. Toxicology findings included the presence of both levamisole and aminorex quantifiable in urine, peripheral blood, brain, liver, and hair. For levamisole, the concentrations were 75.05, 15.05, >0.15, >0.15, and 12.15ng/ml, respectively. The corresponding values for aminorex were 38.62, 8.92, >0.15, >0.15, and 7.35ng/mg, respectively.

Conclusion: This study's results suggest that regular cocaine abusers may be at risk for developing IHP as a consequence of levamisole adulteration, depending on how much levamisole they ingest with their cocaine. Among those who died during the IHP-aminorex epidemic of the early 1970s, the average dose per day ranged from 14 to 42mg and the average decedent had been taking the drug for more than one year. This observation suggests that many current cocaine abusers are at risk. Whether and how many really are at risk cannot be estimated because the conversion rate of levamisole to aminorex in man is not known.

Levamisole, Aminorex, Cocaine

G132 Case Study: Neonatal Postmortem Morphine and Its Pharmacogenetic Implication

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WITHDRAWN

G133 Cause of Death Following Opiate Overdose: A Forensic Epidemiologic Investigation of Comparative Risk

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The goal of this presentation is to familiarize attendees with the concept of comparative risk, a method of causal assessment used in forensic epidemiology, and to demonstrate its application to the investigation of the cause of a death following an alleged act of medical negligence.

This presentation will impact the forensic science community by demonstrating a method of quantifying competing causes of death using epidemiologic data and comparative risk methodology.

An accurate determination of cause of death is central to the resolution of civil and criminal investigations of wrongful death. A fact-finder determination regarding the competing testimony is often reliant, in part, on the qualifications and persuasive abilities of the expert rather than an objective means of comparing the differing opinions. It is well established that a lack of evidence-based practice in forensic medical practice can lead to opinions that rely entirely on an expert's personal medical judgment, which is inherently biased by the experience, or lack thereof, of the clinician with the specific circumstances of a case.

Forensic epidemiology is the subdiscipline of both epidemiology and forensic medicine that is directed at quantifying competing risks of injury or disease that are present in a specific set of circumstances in order to provide an estimate of the likelihood of a specific cause of an injury or disease. The methodology is useful when there are competing causes acting on an individual and, thus, likelihood of causation can be quantified by a metric called "comparative risk." In cases of death following an alleged act of medical negligence, a common competing cause of death is the pre-existing co-morbidity of the patient. The central question in such cases often the "but for" question: But for the exposure to the alleged negligence, what is the chance the individual would have died at the same time?"

Case Presentation: A 55-year-old White male sustained a left distal tibia and proximal fibula fracture in a low-level fall from a ladder. His only significant medical history was of hypertension that was controlled with medication. He was taken to a local hospital and admitted for pain control with intravenous Dilaudid® (hydromorphone), then underwent Open Reduction and Internal Fixation (ORIF) for the fractures. Within the first 22 hours postoperatively, the man also was administered 60mg of OxyContin® and 50mg of regular-release oxycodone, along with temazepam, intravenous morphine, and hydromorphone. Less than 24 hours after the surgery, the man was discharged home, and approximately 10 hours later he was discovered non-responsive at his home, and could not be resuscitated.

Postmortem toxicology results indicated a serum oxycodone level of 0.31 micrograms per milliliter. It was also determined that the man had a dilated cardiomyopathy. The cause of death was ruled "cardiomyopathy and drug intoxication with oxycodone."

The comparative risk question consisted of the risk of death due to opiate intoxication versus the risk of death due to an asymptomatic dilated cardiomyopathy occurring coincidentally at the same time, but unrelated to the opiate exposure. In order to assess the death rate for cardiomyopathy, this study first estimated the cardiomyopathy-related death rate in the sex and age group of the decedent, and compared this to the prevalence of the condition in the same population.¹⁻³ The annual death rate is 12.3 per 100,000

among 237.6 men per 100,000 men of the same age. Thus, 1 in 19.3 men aged 55-64 years old with a diagnosis of cardiomyopathy die in a given year. Therefore, during a ten-hour time frame during the year, the death risk for such a man would be 1 in 16,907 (the annual rate divided by the number of ten-hour periods in the year).

In comparison, the 0.31mg/l of oxycodone is within the range of fatal reactions (lower boundary 0.25mg/l) and more than ten times the level when drug combinations present (lower boundary 0.025 mg/l).⁴ Therefore, while the risk of death due to opiate toxicity in combination with temazepam in a man with cardiomyopathy, is not precisely known, it is certainly greater than 1 in 1,000, and likely greater than 1 in 100. Using the most conservative value, the comparative risk analysis favors the opiate toxicity as the cause of the death by nearly 17 times, versus the cardiomyopathy.

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Forensic Epidemiology, Comparative Risk, Opiate Toxicity



Physical Anthropology



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H1 Accuracy and Reliability of Craniometric Variables Obtained From 3D-Computed Tomography Images

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After attending this presentation, attendees will gain knowledge of two types of measurement error, namely accuracy and reliability, from craniometric variables obtained with Computed Tomography (CT) images.

This presentation will impact the forensic science community by validating the use of CT images as a data source for anthropologists to use in the development or justification of methods employed to estimate a biological profile.

Cranial measurements are routinely used within forensic anthropology and contribute to the key components of the biological profile, including the estimation of sex and ancestry. The measurements are frequently used in the creation of a new method from a skeletal collection or to positively identify an unknown in both forensic casework and mass disaster situations. In any circumstance, obtaining dry skeletal elements requires a time-consuming process inclusive of extensive preparation and processing. Multiple research studies have suggested the use of 3D-CT scans as a tool to collect cranial measurements from the human body, allowing the anthropologist to bypass the need to remove soft tissue in certain situations. In contrast to conventional radiography, the CT scanner produces an anatomical image from thin slices of the object (i.e., cranium) developed from measurements of absorption due to multiple X-Rays made 360 degrees around the object. Although the fundamental design of the CT machine removes the production of distortion, this has yet to be directly compared in the anthropological literature.

A full-body CT scan was performed prior to autopsy at the State of Maryland Office of the Chief Medical Examiner using GE Light Speed RT-16 scanner as well as 3D volume-rendered image generation in the GE Advanced Workstation (AW-2). Following the autopsy, processing the body was conducted to obtain skeletal measurements on dry elements. After all scans and preparation of the remains were complete, 35 standard cranial measurements (both lefts and rights) were collected from the three source types: the dry skeletal elements; the CT images with soft tissue; and the CT images of the dry skeletal elements without soft tissue.

Measurement error has the potential to drastically affect results and subsequent interpretations and thus should be closely evaluated. Because size of the measurement needs to be taken into account, accuracy of the craniometric variables between the three data sources was compared through percent differences. Inter- and intra-observer error was compared through Technical Error of Measurement (TEM), which is the square root of measurement error variance, and relative TEM (%TEM), which allows for a comparison between variables or sources. Agreement

between the measures was also assessed through Bland-Altman plots.

The average percent differences for all source types ranged between 0.6% and 2.0%, indicative of a high accuracy rate. A three-way inter-observer TEM of the cranial measurements obtained on the CT with soft tissue and CT conducted on the dry bone was 2.6mm and 1.78mm, respectively. Three-way intra-observer rates were lower and TEM and %TEM ranged between 0.46 and 0.71 mm and 0.56% and 1.06%, respectively. The intra- and inter-observer rates demonstrate high reliability in landmark location in CT images. Bland-Altman plots illustrate that all measurements are within 2mm from the other two measurements obtained, indicated by all points located within the upper and lower level of agreements.

The results suggest that measurement error associated with craniometric variables obtained on CT images is extremely low and comparable to data collection on dry skeletal elements. More so, the results prove that CT images offer a viable source to obtain metric variables.

Technical Error of Measurement, Percent Differences, Measurement Error

H2 Femoral Midshaft Histomorphometric Patterning: Improving Microscopic Age-at-Death Estimates From Adult Human Skeletal Remains

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After attending this presentation, attendees will understand that the use of microscopic techniques to estimate adult age-at-death is well established within physical anthropology's subfields of bioarchaeology and forensic anthropology. In order to become a more robust technique, however, the long-standing problems of the Osteon Population Density (OPD) asymptote and relatively high Standard Error of the Estimate (SEE) must be overcome.

This presentation will impact the forensic science community by suggesting the posterior Regions of Interest (ROI) be utilized for production of the most accurate and precise microscopic age-at-death estimates from adult human skeletal remains.

Review of the microscopic age-at-death estimation literature reveals that arbitrarily changing skeletal elements, histological variables, sample demographics, and sampling locations have not allowed for accurate age estimation of individuals over approximately 50 years or reduced the standard error of age estimates. This investigation, therefore, began with substantiated theory. All healthy, mobile femurs have in common: genetic programming to establish initial size and shape; the developmental processes of endochondral ossification, appositional growth, and modeling; biomechanical adaptation; periosteal adaptation; cortical thinning and shape change during aging; mechanosensation and mechanotransduction; and bone remodeling.

Building from this theoretical knowledge base, it was

first hypothesized that topographical variation in remodeling exists around human femoral midshaft periosteal cortices that reflects the constraints of normal anatomical development, customary biomechanical usage, and standard mechanobiological functioning. Second, it was hypothesized ROIs associated with the I_{min} second moment of area biomechanical axis would exhibit the lowest remodeling as a result of minimal biomechanical loading. Third, it was hypothesized remodeling at biomechanical ROIs would be histomorphometrically more consistent than at anatomical ROIs due to femoral functional constraints related to obligate striding bipedalism. These hypotheses were tested by counting remodeling events at eight standardized periosteal ROIs (four anatomical—A (anterior), P (posterior), M (medial), L (lateral)—and four biomechanical— I_{maxAnt} , $I_{maxPost}$, I_{minMed} , and I_{minLat}) of 200 adult femoral midshaft cross-sections originally harvested by M.F. Ericksen from George Washington University dissecting room cadavers. The sample was composed of 98 males and 102 females largely of European descent, ranging in age from 30 to 97 years.

While no evidence was found for reduced remodeling at I_{min} ROIs or for more consistent remodeling at biomechanical ROIs, 14 statistically significant differences were found between ROI OPD medians indicating topographical variation in remodeling exists around the femoral midshaft. Specifically, the lowest OPD values occurred at the Anterior ROI, followed by the Posterior, I_{minMed} , $I_{maxPost}$, I_{maxAnt} , I_{minLat} , Medial, and Lateral ROIs. Additionally, although the anterior femoral cortex has traditionally been sampled for microscopic age-at-death estimation, here, the Anterior ROI was found to reach the OPD asymptote at approximately 50 years of age and was associated with the highest SEE. Alternatively, the Posterior ROI, the location possessing the second-lowest median OPD value, was found to be associated with the lowest SEE and showed no sign of having reached the OPD asymptote. It therefore bears further investigation to see if this pattern can be replicated across multiple samples.

Histomorphometrics, Age-at-Death, Regions of Interest

H3 The Use of Micro-Computed Tomography to Determine Volumetric Shrinkage Trends of Thermally Damaged Bone

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After attending this presentation, attendees will gain an understanding of the potential of micro-Computed Tomography for the advancement of forensic anthropological research as a nondestructive means of measuring changes in the bones' micro-architecture, volume, and stereology.

This presentation will impact the forensic science community by demonstrating the usage of micro-CT to accurately document and quantify shrinkage as well as morphological changes of heat-exposed bone, allowing a more in-depth comprehension of bone's structural changes in response to heat and its implications for techniques of biological profile estimation.

In recent years, micro-CT has become an emerging practice in the forensic sciences. Its application in forensic

anthropology has, however, to date been minimal despite its undeniable potential. Due to the high signal contrast between bone and soft tissue and its significantly higher spatial resolution than standard clinical CTs, the technology is already finding frequent use in medical pre-clinical investigations. In forensic settings, micro-CT has been utilized to determine class and individual characteristics of tool marks on bone, and its application toward the visualization of age-related changes of bone landmarks such as the pubis or sternal rib ends, allowing for a quantification of parameters such as ridge depth and surface areas without the removal of soft tissues, has been discussed in the literature.¹

The present study explores the usage of micro-CT to assess and quantify changes in the volume and microstructure undergone by bone subjected to heating. The phenomenon of thermally induced shrinkage and the implications for forensic anthropology, particularly the effect on metrical techniques such as size and stature estimation, have been discussed and investigated by a number of researchers over the past decade. Studies have reported vast discrepancies in bone shrinkage, ranging between 1% and 27% depending on factors such as the experimental conditions, the skeletal element and its compact to spongy bone ratio, mineral composition, and fragment size.² However, all of these observations are based on manual caliper measurements, not taking into account the three-dimensional volumetric shrinkage and changes in bone micro-architecture and stereology which would allow for a more accurate determination of absolute shrinkage. To date there has solely been one study carried out using micro-CT to investigate the heat-induced shrinkage of dental tissues, showing the feasibility of this approach.³

Research was carried out using fresh de-fleshed sheep ribs (*Ovis aries*) cut into 0.5cm - large sections and burned in an ashing furnace in triplicates at temperatures between 400°C (the average temperature of a camp fire) and 1000°C (average temperature of a crematorium) in 100°C increments for 45 minutes. All samples were scanned pre- and post- burning using a SkyScan™ 1172 micro-CT scanner at 13.59µm resolution using 49kV voltage, 120µA current and a 0.5mm aluminum filter. The resulting files were subsequently reconstructed using the SkyScan's™ NSRECON package utilizing a uniform attenuation coefficient.

Pearson's correlation coefficient shows a clear correlation between temperature and degree of shrinkage at the 0.01 level ($R^2 = 0.905$). Average volumetric shrinkage ranged from 14.0% (at 400°C) to 45.5% (at 1000°C), which corresponds to 5.8% to 12.7% shrinkage in the diameter. ANOVA with post hoc LSD test showed no significant difference in shrinkage at temperatures up to 600°C ($p > 0.05$). An increase in shrinkage was found in the 700°C and 80°C samples ($p < 0.05$). Bones burnt at temperatures of 900°C and 1,000°C both showed significantly higher shrinkage than any other temperature groups ($p < 0.05$). Further analysis of the dependency of shrinkage on factors such as fragment size, compact thickness and the compact-to-spongy bone ratio are planned.

The findings of this research signify the vast potential of micro-CT in forensic anthropological research on the effects of factors such as diagenesis or burning on the bones density, morphology and micro-architecture, a comprehensive understanding of which is crucial for the correct interpretation of biological profile and taphonomy of skeletal remains in a forensic context.

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Micro-CT, Cremains, Fire Victim Identification

H4 Trace Element Analysis of Human Tooth Enamel by LA-ICP-MS for Estimating Region of Origin

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After attending this presentation, attendees will have an understanding of the geologic and anthropogenic origins of trace elements in tooth enamel as well as their utility in estimating region of origin. A method of non-matrix-matched calibration for analyzing human tooth enamel using Laser Ablation-Inductively Coupled Plasma-Mass Spectrometry (LA-ICP-MS) will also be described.

This presentation will impact the forensic science community by contributing to new techniques for estimating the region of origin of unidentified individuals, which may increase the likelihood of identification. It may be particularly important in mass fatality situations.

Tooth enamel is among the most durable substances in the human body and as such has high recoverability in forensic anthropology cases. Its crystalline hydroxyapatite matrix, though primarily composed of calcium, has a slightly variable chemical composition in which substitutions are the result of biologically available trace elements. The trace elements are derived from an individual's diet and the water they consume during the period of enamel formation. Enamel formation of permanent teeth occurs mostly during early childhood, though crown formation of the third molars is not complete until late adolescence. Thus, the trace elements present in an individual's permanent teeth reflect the geology of the area in which they resided during this period; for fairly sedentary populations, this can be assumed to approximate an individual's birthplace. Anthropogenic sources of trace element uptake such as imported foods, contaminants from cookware, and dental restorations must also be considered. Once anthropogenic sources and natural variation in the elemental composition of tooth enamel are accounted for, the remaining variation can be used to distinguish teeth originating from different individuals, and may be useful in estimating an individual's birthplace.

This research examines a sample of teeth from the Antioquia Modern Skeletal Reference Collection in Medellin, Colombia. The sample has known demographic information including sex, birthplace, and age. For this study, 71 teeth from 61 individuals from areas throughout northwestern Colombia were analyzed; 36 of these individuals were born in the city of Medellin, while the remaining 25 were born in surrounding areas. This sample includes 54 males and 7 females with a mean age-at-death of 35 years and a standard deviation of 17 years. It consists of an assortment of types of teeth based on availability in the collection and ease of extraction. The teeth were analyzed LA-ICP-MS, a minimally destructive, semi-quantitative technique. The exterior surfaces of whole and fragmented teeth were used. LA-ICP-MS analysis was performed at the Center for Archaeology, Materials, and Applied Spectroscopy at Idaho State University.

Due to the nature of LA-ICP-MS analysis, quantitation

requires the use of matrix-matched standards to calibrate for the effects of the matrix on the instrument. Since no matrix-matched standard is available for human tooth enamel, Scanning Electron Microscopy-Energy Dispersive Spectrometry (SEM-EDS) was utilized for non-matrix-matched calibration. This allows for semi-quantitation of the elemental concentrations in the sample. A suite of up to 44 elements was examined: sixteen of these show variation across the sample and six (Li, Al, Cu, Sr, Sn, Ba) may potentially be useful for estimating region of origin. For example, among the individuals born in Medellin, aluminum has an average concentration of 2 ppm and a standard deviation of 2ppm; for the other individuals, the mean is 4ppm with a standard deviation of 8ppm. Barium has a mean concentration of 4ppm and a standard deviation of 2ppm among the Medellin subsample, while among the remaining individuals the mean concentration is 14ppm with a standard deviation of 39ppm. This illustrates that the 36 individuals born in Medellin have a more similar compositional profile than do those born in other areas. These results suggest that trace element analysis of permanent tooth enamel may be of some use in estimating an individual's region of origin in forensic anthropological contexts.

Tooth Enamel, Region of Origin, LA-ICP-MS

H5 Inter- and Intra-Observer Reliability Using the Walker Non-Metric Sexing Technique

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After attending this presentation, attendees will better understand observer error rates using non-metric cranial traits and the effects of observer error when estimating the sex of unknown individuals.

This presentation will impact the forensic science community by providing knowledge on the reliability of using non-metric cranial sex traits and the practical consequences of observer error when estimating the sex of unknown skeletal remains in a forensic context.

While the validity of traditional non-metric techniques of sex estimation has been contested due to their subjective nature, some studies have found that certain non-metric traits do perform well when examined by multiple observers.^{1,2} A study by Walker evaluated the use of five non-metric traits for sex estimation and provided descriptions and illustrations to demonstrate specific levels of expression for each trait.^{3,4} The inter- and intra-observer error rates were calculated and the assigned scores between observers were found to fall within one score of the modal value 96% of the time and 99.5% for the intra-observer test.³ These high levels of agreement suggest that the method can be reliably used. As the method is commonly used to analyze forensic cases, it is important to validate the reliability of the method. This study was designed to test the accuracy of Walker's reported inter- and intra-observer rates for each cranial trait. In addition, the practicality of this method was examined through the consequences of disagreement between and within observers in an effort to identify traits that need better clarification and description.

The Walker method was used to score five cranial traits in African American individuals from the Hamann-Todd Collection (total n=59). Three different observers scored the crania, two of whom scored the crania twice with at least a week between scoring rounds. Statistical analyses were performed to calculate the inter- and intra-observer error rates. The scores were also applied to the equation provided by Walker with the highest reported accuracy

rates. The estimated sexes obtained were then compared across observers and to the known documented sex to evaluate the forensic consequences of observer error.

Pairwise comparisons completed using Cohen's weighted kappa revealed that there was moderate to substantial agreement (0.43-0.80) between and within observers for the mastoid process, orbital margin, and glabella, fair to substantial agreement (0.25-0.64) for the nuchal crest, and only slight to fair agreement (0.05-0.38) for the mental eminence. The percentage of agreement tests between observers indicated that the mastoid process, orbital margin, and glabella were the most consistently scored traits, while the mental eminence performed the worst with agreement ranging from 52.5% to 67.8%. Similarly, the intra-class correlation coefficient tests demonstrated that deviations between observers' scores for each round were greatest in the mental eminence with low agreement scores (0.07 and 0.20). For all tests, glabella performed best with high levels of agreement while mastoid and orbital margin exhibited moderate levels of agreement. The correct classification rates for each observer ranged from 67.8% to 79.7%, which are lower than Walker's published rates.³ Overall, the results indicate that accuracy rates of this method are affected by observer error of certain traits which could have a vital impact in the forensic application of this method.

Results show that the mental eminence is the least reliable cranial trait for sex determination both between and within observers. The mental eminence had the lowest agreement scores for all inter- and intra-observer error tests followed by the nuchal crest scores; however, the equation used from the original study by Walker suggested to have the highest accuracy rates gives the mental eminence approximately equal weight as the glabella and mastoid process scores.³ While the mental eminence did not prove to be a reliable trait, glabella and the mastoid process had high levels of agreement which likely enhances the efficacy of the method. The low reliability of the mental eminence could either be the result of low levels of sexual dimorphism or it may reflect the need for enhanced visual and descriptive tools to capture the sex difference in shape and more objectively score this feature. Until then, more reliance should be placed on methods utilizing the glabella, orbital, and mastoid regions as these traits proved to be reliably scored cranial traits both between and within observers.

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Observer Error, Sex Estimation, Non-Metric

H6 Provenancing Human Remains From Forensic Contexts: Application of Stable Isotope Analysis in Forensic Anthropology

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The goal of this presentation is to highlight the role of stable isotope analysis as an investigative tool for provenancing human remains to aid in narrowing search parameters for missing persons. After attending this presentation, attendees will understand how stable isotope analysis can be used in conjunction with the biological profile, especially in situations where traditional methods have not been successful.

This presentation will impact the forensic science community by illustrating case studies in which stable isotope analysis has aided in the identification process.

Stable isotope analysis of human tissues can provide contextual information on the diet and migration history of individuals. In this study, stable isotope analysis was conducted to determine whether two individuals were likely local or nonlocal to the area where the remains were discovered.

The first case, recovered from a dry creek bottom in the Central Valley of California, consisted of surface-scattered human remains representing a partial skeleton. The biological profile suggests the individual was an adult male. Ancestry could not be estimated due to absence of the cranium. Stable carbon isotope values are -14.3‰ for bone collagen and -10.0‰ for bioapatite, indicating the decedent consumed a diet consisting of relatively equal amounts of C3 and C4 resources. The stable nitrogen isotope value of 11.2‰ is consistent with an omnivorous diet. Both stable carbon and nitrogen isotope values of bone collagen are significantly elevated relative to published data on modern hair keratin of U.S. Americans ($\delta^{13}C$: $z = 3.6$; $\delta^{15}N$: $z = 5.8$), suggesting a diet more typical of individuals from Latin America, where corn (a C4 plant) is a common dietary staple.^{1,2} Furthermore, the stable oxygen isotope value of -6.4‰ is not consistent with local tap water sources, suggesting the decedent was nonlocal to the area where his remains were found. The $\delta^{18}O$ value is consistent with tap water sources along the eastern border of the Central Valley, and certain regions of the Southwestern U.S. and Mexico. DNA testing later confirmed the identity of the decedent, a 37-year old Hispanic male, from Tepatitlan de Morelos, Jalisco, Mexico, who had moved to northern California prior to his death. The $\delta^{18}O$ value is consistent with this region of Mexico, but is not consistent with his most recent place of residence.

The second case consists of an isolated mandible recovered from the North Coast Range of northern California. Morphological and metric assessments suggest the decedent was an adult male, although classification for ancestry was indeterminate. Stable carbon isotope values are -19.0‰ for bone collagen and -13.4‰ for bioapatite, indicating the decedent primarily consumed C3 food resources with little input from C4 resources. The stable nitrogen isotope value of 11.0‰ suggests an omnivorous diet. Although stable carbon and nitrogen isotope values of bone collagen are also significantly different from published data on modern U.S. Americans ($\delta^{13}C$: $z = 2.25$; $\delta^{15}N$: $z = 5.3$), little comparative modern dietary data exist for this area of California.^{1,2} The stable oxygen isotope value of -7.9‰ is consistent with local tap water sources in the region, suggesting the decedent may be local to the area. These data support a tentative identification of a missing male who resided in the area for his entire life. Future isotopic analysis of a mandibular molar may further refine the geographical parameters.

These case studies reveal examples of valuable contextual information provided by stable isotope analysis that can assist in identification efforts. Stable isotope analysis can be implemented to support a proposed identification, as in case 1, or to narrow the scope of possible missing persons to consider, as in case 2. The benefits demonstrated in these cases support the continued use of stable isotope forensics in conjunction with other biological profile assessments.

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Forensic Anthropology, Stable Isotopes, Human Provenancing

H7 The Utility of Baking Bone to Increase Skeletal DNA Yield

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After viewing this presentation, attendees will understand whether artificially baking bones will increase skeletal DNA yields obtained during the skeletal DNA extraction process and whether baking bones impacts the quality of the DNA required for subsequent genetic analysis.

This presentation will impact the forensic science community by contributing information on a new technique aimed at increasing DNA yields obtained during the skeletal DNA extraction process. The ability to increase skeletal DNA yield by artificially heating the bone may provide the analyst with a greater DNA yield for subsequent genetic analyses and identification efforts.

It is well established that DNA can be extracted from both soft and hard tissues. Once the body dies the cells and cellular components, including the DNA, begin to degrade. The body's soft tissues will decompose at a faster rate than the osseous tissues resulting, in many cases, in the skeleton being the only biological material remaining at the crime scene. For this reason, the skeleton is an alluring target for genetic analyses aimed at individual identification. However, isolating DNA from osseous tissue can be challenging. On the one hand, bone affords some protection to the skeletal DNA by providing a physical barrier to environmental agents that could potentially act to degrade the molecular material. On the other hand, the skeletal DNA becomes chemically bound to the inorganic portion of the bone and this chemical modification makes skeletal DNA more difficult to purify than soft tissue DNA. There is a growing body of research aimed at improving DNA yields obtained during the skeletal extraction process. While heat tends to negatively impact biological preservation at both the gross and molecular levels, recent information suggests heating bone may be beneficial in increasing skeletal DNA yields by making the osseous material more brittle and therefore better able to release the DNA bound to the mineral phase. However, this hypothesis had never been systematically investigated until now.

This research utilized a sample of seven dry (unmacerated) pig femora of adult or near adult age collected from the ground surface of an Indiana farm. Ten small 1x1cm squares of cortical bone were cut from each shaft, resulting in a final sample size of 70 skeletal DNA extractions. Nine samples from each individual were baked in a mechanical airflow oven at 90°C for different increments of time (3, 6, 9, 12, 18, 24, 36, 48, and 72 hours). The last segment of each bone served as an unbaked control to standardize for possible variation in the starting levels of DNA. After extraction, the DNA yields were quantified and the extracted DNA was Polymerase Chain Reaction amplified to determine DNA quality. PCR amplification employed pig-specific DNA primers that target nuclear Short Tandem Repeats (STRs). Pig STRs were targeted primarily

because they span a region that consists of a variable number of repeating units similar to the human STRs that are routinely used in forensic DNA analysis. Several null hypotheses were tested: (1) DNA yield from baked bones will not differ from those that are unbaked; (2) DNA yield is not correlated with accumulated heat as measured by Accumulated Degree Hours (ADH); and, (3) PCR success is not dependent on accumulated heat.

Results indicate that artificially heating bones by baking, on average, increases skeletal DNA yield compared to the unheated controls. A *t*-test was used to determine whether yield from baked bones differed from the unbaked sample and was found to be significant ($t = -3.81, p = 0.027$). After plotting the mean yields versus time, a pattern of increasing yields was evident as heating time increased. A Pearson's correlation between quantity of extracted DNA and ADH was significant as well ($r = 0.298, p = 0.016$). However, a drop-off point at which heating became detrimental was not evident. Lastly, a Spearman's correlation used to test whether there was an association between heating and PCR success was not significant. This research has found that there may be some benefit in baking bones before skeletal DNA extraction.

Skeletal DNA, Bone Baking, DNA Yield

H8 Mapping Spatial Patterns in Cortical Bone Histology From the Femoral Midshaft Using Geographic Information Systems Software

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After attending this presentation, attendees will be better able to conceptualize the spatial variation and distribution in skeletal remodeling events across the entire cortex of the femoral midshaft.

This presentation will impact the forensic science community by demonstrating the use of Geographic Information Systems (GIS) software as a new method for the analysis of bone microstructure that may better inform sampling procedures employed for the estimation of age-at-death through histological means.

When constructing a biological profile of unknown human skeletal remains, forensic anthropologists must estimate the age-at-death of the decedent. As a complement to traditional macroscopic methods employed, or when the necessary macroscopic elements are absent, age estimation can be conducted through a histological examination of remodeling events in cortical bone. During the last half century, the femoral midshaft has been the most commonly employed skeletal site for obtaining histological age estimates; however, different methods employ various sampling locations that differ in size, number, and location for the collection of histological data across the cortex. Building upon recent work that has demonstrated the utility of GIS software for the study of human bone microstructure, this pilot study employed GIS software to examine patterns in the spatial distribution of histological remodeling events across the entirety of the femoral midshaft.¹

Ten femoral midshaft samples from modern cadaveric donors (five male, five female) were photographed under polarized light and micrographs were compiled into seamless cross-sectional images; samples ranged in age from 30-76 years at time of death, with a mean age of 54.5 years. Images were imported into ArcGIS™ (v. 10.1), where polygon features were created to overlay the cortical area and point features were created to mark all intact and fragmentary osteons, as well as resorption spaces. A total of

66,491 remodeling events were manually notated and density maps were generated to visualize the clustering of remodeling events across the cortex. Density maps revealed a striking qualitative pattern, with the highest density of remodeling events in the lateral/ anterolateral cortex across both sexes and all ages.

To quantitatively analyze this pattern, cortical areas were digitally divided into quadrants to delineate standard anterior, medial, posterior, and lateral quadrants, as well as offset anterolateral, anteromedial, posteromedial, and posterolateral quadrants. The total number of remodeling events contained within the area of each quadrant was used to determine Osteon Population Density (OPD; remodeling events/mm²). One-way Analysis Of Variance (ANOVA) tests demonstrated a significant difference in OPD values for both the standard quadrant system ($F=5.752, p<0.01$) and the offset quadrant system ($F=6.031, p<0.01$). A statistically significant positive correlation coefficient was found between OPD and age-at-death ($p<0.05$) for all standard and offset quadrants with the sole exception of the posterior quadrant, which was not statistically significant.

Remodeling events were further investigated for their distribution across the depth of the femoral cortex. Measurements were taken to digitally divide the cortex into periosteal, middle, and endosteal cortical thirds. A one-way ANOVA test yielded no significant difference between OPD values for the periosteal, middle, and endosteal thirds ($F=2.726, p>0.05$). However, correlation between OPD and age differed greatly across the depth of the cortex: the periosteal third yielded the highest correlation ($r=0.911, p<0.001$); the middle third was also significant ($r=0.729, p<0.05$); and the endosteal third did not significantly correlate ($r=0.600, p>0.05$). A significant correlation was also found between OPD and age for the entire femoral cortex ($r=0.854, p<0.01$).

This pilot study demonstrates the utility and potential of using GIS software for mapping spatial patterns of histological remodeling in human cortical bone. Continued research is underway to further refine which sampling location (or combination of sampling locations) across the cortex will yield the best age-at-death estimates, with specific focus on determining when different cortical regions become saturated with remodeling events and reach the OPD asymptote. Elucidating these patterns will be of great use to forensic anthropologists who use histological methods for estimation of age-at-death from human skeletal remains.

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Bone Histology, Osteon Distribution, Bone Mapping

H9 Standardized Descriptive Method for the Anthropological Evaluation of Pediatric Skull Fractures

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The goal of this presentation is to provide case example illustrations of a novel method developed for the standardization of pediatric skull fracture documentation.

This presentation will impact the forensic science community by describing a method designed for use by anthropologists in the description of pediatric skull fractures, and thus circumvents the confounding effects of the inappropriate application of clinical fracture descriptions in the forensic context.

Distinguishing accidental from non-accidental head trauma is an important component of the forensic autopsy and is based on the thorough detection, standardized description, and appropriate interpretation of skull fractures. The majority of the literature pertaining to pediatric skull fracture is clinically based and, thus, motivated by the need for effective assessment of both fracture characteristics (type, frequency, location, and mechanics) and context (severity of injury, associated soft tissue damage, and patient prognosis). This clinical perspective is reflected in the significant array of skull fracture classification systems that exist. Most clinical schema distinguish some variation of the following categories: simple, complex, comminuted/composite, depressed, basilar, stellate, diastatic, growing, and ping-pong. From a strictly descriptive standpoint, these categories are not mutually exclusive and instead represent overlapping levels of detail in fracture description, which are useful in the clinical setting but confound the non-clinical description of fractures in the forensic context. For this reason, application of these schema in the forensic anthropological interpretation of skull fractures, as manifest on the bone itself rather than imaging/associated clinical findings, is inappropriate.

It is argued that forensic anthropological interpretation of skull fractures requires a standard classification system that reflects fracture morphology alone, and a three-stepped classification system which conveys increasing detail with each additional step is suggested. The first and most basic step is the fracture *category* of which three variations exist: simple, complex, and comminuted. The second step describes the fracture *pattern*, and the third step adds the fracture *descriptors*. Thus, the proposed system utilizes the fracture characteristics that covary with, but are not independent, of the basic fracture categories, as modifiers rather than additional fracture categories.

The proposed schema was applied retrospectively to Harris County Institute of Forensic Sciences (HCIFS) anthropology cases analyzed between April 2007 and April 2012 to evaluate its applicability and repeatability. The study sample included all children age two years and under for which an anthropological examination was requested and that had at least one skull fracture.

The study was intended to statistically evaluate: (1) the effectiveness of the method in capturing the variation in pediatric cranial fractures seen at the HCIFS, and, (2) to identify the potential for interobserver error in the application of the schema. For each case, the diagrams and handwritten notes completed by the examining anthropologist, autopsy, anthropology photographs, and micrographs were reviewed.

Forty-four skull fractures were found in the 31 cases in the sample. Application of the method demonstrated that the proposed nomenclature adequately captured and concisely recorded fracture morphology variation in each of the cases and validated the effectiveness of the schema in describing pediatric cranial fractures. There was 100% agreement between the four anthropologists in the assignment of the fracture category (simple, complex, or comminuted). The fracture patterns assigned to each fracture were also highly consistent among the analysts, with 79% agreement between observers. The discordant entries did not represent contradictory fracture pattern interpretation but rather slight interpretational variation.

This study illustrates the value of the proposed nomenclature for the standardization of the anthropological description of skull fractures and the effective distinction of fracture description and clinical implication. The system is adequate for the anthropological classification of the majority of skull fractures observed during medicolegal autopsies of children two years of age

and younger. This presentation will outline the practical application of the method using HCIFS case examples.

Child Abuse, Skull Fracture, Blunt Trauma

H10 Craniometric Concordance: 3D Surface Scanner and Digitizer Measurements

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After attending this presentation, attendees will better understand the level of agreement between human cranial measurements derived from 3D digitizer and 3D surface scanner techniques as well as the implications of using these differently derived measurements for sex and ancestry estimation.

This presentation will impact the forensic science community by providing information on which cranial measurements demonstrate the greatest variability between the two techniques, focusing specifically on measurements used for sex and ancestry estimation in FORDISC® 3.1. Additionally, the practical effects of measurement differences on discriminant function analyses will be examined in regard to the validity of sex and ancestry estimations in forensic cases.

Forensic anthropologists have historically relied upon caliper measurements and, more recently, 3D digitizer measurements taken from the cranium to estimate the sex and ancestry of unknown skeletal remains. Recent advances in technology have provided practitioners with new resources for collecting data from human crania. For example, 3D surface scanners have become an important tool because of their ability to create virtual models of human crania that can be archived. From these three-dimensional models, morphological features can easily be extracted and analyzed using engineering and morphometric software. Interlandmark distances can also be easily obtained and are assumed to be comparable to traditional osteometric measurements. Although previous studies have been conducted to assess the accuracy of 3D surface scans and the ability of researchers to locate traditional osteometric landmarks, sample sizes have been limited and none have attempted to analyze the differences between digitized and virtual measurements from a multivariate perspective for classification purposes.

This study analyzed 15 cranial measurements on a pooled sample of human crania (n=70), representing approximately equal numbers of 19th- and 20th-century U.S. Black and White males and females from the Robert J. Terry Anatomical Skeletal Collection. Osteometric landmark coordinates were collected from each cranium using a MicroScribe® 3D digitizer. A NextEngine® desktop 3D laser scanner was used to collect 3D surface scans from the same crania. Cranial measurements were then calculated from the surface scans using Geomagic Studio 12. Notably, the initial digitization occurred many years before scanning and virtual measurement, and all three activities were conducted by three different individuals. The digitized and scan measurements from the 70 crania were then treated as paired data sets and univariately and multivariately compared to assess the level of agreement between the measurements derived from each method.

Paired *t*-tests detected significant differences ($p < 0.05$) between the means of the 3D scanner and 3D digitizer measurements for ten of the fifteen cranial measurements. The mean differences ranged from 0.01 to 1.26mm and the standard deviation of the differences ranged between 0.85 and 3.05mm. The coefficient of variation for differences in each measurement

ranged from 0.71 to 6.47. Bias was not consistently positive or negative throughout the fifteen measurements; measurements taken from the 3D surface scans resulted in positive bias for seven measurements and negative bias for the remaining eight. The samples were also subjected to discriminant function analysis in FORDISC® 3.1 to classify using measurement technique.¹ Using forward mean stepwise selection, the highest total correct classification rate obtained was 67.4%. When the two samples were analyzed against a third group of 70 crania (20th-century Black males in the Forensic Data Bank), the Mahalanobis distances for all group comparisons were significant ($p < 0.01$), except when comparing the 3D digitizer and 3D scanner groups ($p = 0.031$). This indicates an inability to truly separate the samples based on measurement differences, suggesting that the two methods are comparable.

This research suggests that while caution should always be exercised when attempting to combine data obtained from these two techniques, measurement differences may be less significant than previously thought and the use of both methods may be applicable in forensic contexts.

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Cranial Measurements, Digitizer, Surface Scanner

H11 A Comparison of Metric and Non-Metric Techniques Used in the Classification of Hispanic Crania

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The goal of this presentation is to evaluate and compare standard metric and non-metric techniques commonly used in the classification of Hispanic crania. Attendees will have an increased knowledge of the applicability and accuracy of various metric and non-metric techniques in the determination of Hispanic ancestry.

This presentation will impact the forensic science community by illustrating the issues and complexities involved in accurate identification of Hispanic crania. It highlights the utility, importance, and need for correlation of both metric and non-metric data in the successful classification of these hybrid crania.

In the post-*Daubert* forensic community, there has been a push to abandon more traditional, experience-based methods in favor of those that are more statistically rigorous and replicable. While this is of utmost importance in terms of achieving reliable results that affect the outcomes of legal situations, there may be situations in which methods based on experience may prove their worth. One such instance where this is evident is the attribution of ancestry to cranial remains of Hispanic decedents. The difficulty of successfully classifying Hispanic crania has previously been noted.¹ Because Hispanic individuals can be culturally and genetically heterogeneous admixtures of Native Americans, African Blacks, and European Whites, trait expression varies and is often intermediate between extremes. This Presentation aims to evaluate morphological traits associated with Hispanic crania and compare their success in classification to metric trait analyses to determine which mode of analysis is able to better identify a sample of Hispanic crania.

A blind sample of 50 male and female crania from the William M. Bass Donated Skeletal Collection was utilized in this study. Metric analysis of these crania consisted of discriminant function analysis through the use of FORDISC® 3.1 using a standard

suite of 24 cranial measurements.² Crania were also digitized and classifications achieved using Geometric Morphometric (GM) methods. Nonmetric traits recorded from each cranium followed those described by Birkby *et al.*, Hurst, and Peacock and Zinni and included estimation of alveolar prognathism, nasal overgrowth, nasal aperture width, nasal spine development, supranasal suture morphology, eye orbit shape, shoveling of the incisors, as well as morphology of the frontal process of the zygomatic.³⁻⁵

The use of non-metric traits resulted in correct classification of nearly 77% of the Bass Hispanic crania. Using all-group classifications, FORDISC® 3.1 was able to correctly classify 31% of the Hispanic sample; however, when Mahalanobis distance matrices and canonical variates plots were considered, successful classification rates improved significantly. GM analysis of Bass crania, when combined with non-metric assessments, provided the most accurate Hispanic crania classification rate. These results illustrate that although metric methods for Hispanic ancestry classification can be applied more objectively and replicated more easily in forensic anthropology, correlation of morphological (i.e., nonmetric) methods with metric methods can potentially provide results not attainable by either used alone.

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Hispanic, Crania, Ancestry

H12 Positive Identification Using Panoramic Radiographs: A Visual Assessment of the Maxillary Anatomical Structures

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After attending this presentation, attendees will gain an understanding of a method of decedent identification using comparison of the maxillary region of antemortem and postmortem panoramic dental radiographs.

This presentation will impact the forensic science community by providing forensic anthropologists with an additional method of making a positive identification of an unknown individual.

Positive identification of unknown decedents is of great importance in any investigation. This process is often made difficult by a lack of antemortem records for comparison. A variety of positive identification methods using radiographic comparison of various anatomical structures have been extensively studied to combat this limitation, but many of these methods focus on comparisons of radiographs that are not common in antemortem records and can only be used in isolated cases. Standard dental radiographs are commonly used to make positive identifications of unknown remains because of their availability in antemortem records, but these identifications focus on the dentition of the individual. In recent years, panoramic dental radiography, which allows for the examination of both dentition and a large portion of the maxillary

and mandibular regions, has become commonly used in dental practices across the United States due to the increased diagnostic ability they provide. These panoramic dental radiographs have the benefit of being relatively common in the antemortem record, and could be used to compare a number of anatomical structures within the facial region of the cranium if dental identification was not successful. However, the use of these structures in the positive identification process has not been fully researched.

The purpose of this pilot study was to examine the maxillary region of a known sample of pairs of panoramic radiographs from 45 individuals, looking specifically at the maxillary sinus region visible on the radiographs for both congruency and corresponding unique traits between the pairs of radiographs. This sinus region was chosen due to the previous success of studies on positive identification using radiographs of the frontal sinuses. To examine this, the dentition was cropped out of each radiograph, and an online survey was created that showed radiographs one at a time and allowed for the selection of a match for the radiograph from a group of five possible radiographs. Twenty participants completed both the radiograph matching survey and a brief questionnaire noting qualifications and level of experience. In addition to selecting matches for the radiographs, the participants were also asked to select points on the chosen radiograph match that were diagnostic for them in making their selection.

The results indicated that the maxillary region of panoramic radiographs can be matched with an average of 76% accuracy, indicating moderate success in positive identification. Accuracy for matching the radiographs was significantly higher for the participants with more experience. Four anatomical structures were most associated with being diagnostic of a positive match: the laterobasal border of the nasal cavity, the inferior nasal conchae, the borders of the eye orbits, and the maxillary sinuses. These results indicate that the maxillary region of panoramic dental radiographs has the potential to be used for positive identification purposes in the field of forensic anthropology and should be pursued further due to high inclusion of panoramic radiographs in the antemortem record. Future studies should further evaluate this region with both geometric shape analyses and visual matching assessments.

Radiographs, Positive Identification, Comparison

H13 Study of the Sexual Dimorphism of the Postcranial Base With MSCT

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The goal of this presentation is to evaluate the sexual dimorphism of the foramen magnum and the occipital condyles using geometric morphometrics on Multislice Computed Tomography (MSCT).

This presentation will impact the forensic science community by providing an example of an anthropological application of the Multislice Computed Tomography in forensic sciences.

MSCT is uncommonly used in forensic anthropology.

MSCT allows more precision than metric and scopic methods, which can be helpful for sex determination.

The technique: 120 angio CT scans from the radiology department of the university hospital of Toulouse (France) with a sixteen-detector row MSCT (Sensation 16, Siemens) were selected. For each angio CT scan, size and shape of the postcranial base were studied.

To study the size of the postcranial base, 20 landmarks, based on literature data, were positioned on Amira® 5.2.2: four landmarks on the foramen magnum to appreciate length and width; four landmarks on each occipital condyle to appreciate length and width; four landmarks on the Frankfort plan; and four landmarks on the maximum length and width of the skull to appreciate the ratio between foramen magnum and skull measurements.

To study the shape, outlines of the foramen magnum were obtained using ImageJ® from snapshots of each foramen magnum realized on Amira® 5.2.2. With each outline, two processes were used: Fourier analysis to obtain 3D statistical shape models, using geometric morphometrics, via R 2.1.5 software, and shape descriptors using form factors (perimeter, area, roundness, compacity, aspect ratio, and circularity). For each measurement (size and shape) with R 2.1.5 software, mean, standard deviation, median, maximum and minimum data were calculated using boxplots. The sample studied consisted of 60 males and 60 females.

Study of Size: The width of the foramen magnum was higher for men than women, as noted in literature, but it was not a statistically significant result, as previous studies showed. The length of the foramen magnum was higher for men and it is a statistically significant result. Men have higher occipital condyles than women, whether it is the left or the right condyle, which is statistically significant. There is no statistically significant difference between men and women concerning the angle formed by the foramen magnum and the Frankfurt plan, as shown on literature data. There is no statistically significant difference between men and women concerning a ratio between the length of the foramen magnum and the maximum length of the skull. The width ratio between the foramen magnum and the skull is higher for women, which is a statistically significant result.

Study of Shape: From the Fourier analysis, there is a superposition of the points concerning men and women on the PCA, which proves an absence of clear discrimination between men and women. The results are statistically significant for only two shape descriptors (area and perimeter).

This study is the third study performed on MSCT about foramen magnum sexual dimorphism; the most previous studies concerned dry bone. The length of the foramen magnum is a statistically significant measurement, but not its width. This information is new and differs from the literature, which shows the opposite. The right occipital condyle (using length and width) is discriminant, as shown in the literature, but this study shows that the left occipital condyle is discriminant too. Other new information was obtained by studying the ratio between measurements of the foramen magnum and those of the skull. It was shown that women have higher ratio between width of the foramen magnum and the maximum width of the skull than men. This unpublished result is statistically significant and shows the existence of a morphological criterion.

Generally, there is no sexual dimorphism for the shape of the foramen magnum. Shape descriptors are not a pertinent tool to study foramen magnum sexual dimorphism. This study shows a robustness criterion (length) because of a sexual dimorphism on the size of the foramen magnum and a morphological criterion (width ratio between foramen magnum and skull), but no shape effect.

Anthropology, Sexual Dimorphism, Tomography

H14 The Use of Osteometric Sorting Techniques to Aid in the Resolution of Large-Scale Commingling

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After attending this presentation, attendees will understand how anthropological techniques can be used to aid in the resolution of large-scale commingling of human skeletal remains. The gross and metric sorting procedures can be beneficial in cases of large-scale commingling, particularly in situations when there are time and monetary constraints.

This presentation will impact the forensic science community by demonstrating the utility of gross and osteometric sorting techniques in the investigation of large-scale commingling such as in post-conflict and mass disasters.

The purpose of this study was to illustrate the practicality of utilizing gross and osteometric sorting techniques as a first approach in the sorting of commingled human remains. The Piggot archaeological ossuary site (31CR14), curated at the Forensic Analysis Laboratory at North Carolina State University was employed to represent a large-scale commingling. The Minimum Number of Individuals (MNI) was estimated at 78. The MNI was calculated using the right humerus, which was the skeletal element and side most represented in the assemblage.¹

The sample used in this study consists of 114 skeletal elements. Each individual element was assigned an identification number. All bones were measured according to standard and non-standard measurements and recorded in a Microsoft® Excel® spreadsheet.²⁻⁴ In cases of fragmentation, only available measurements were taken. All *t*-distributions were performed in Excel®; regression equations were derived in SPSS® Statistics 19.0.0.

Visual pair matching, the association of left and right elements based on parallels in morphology, was conducted on all complete or nearly complete elements.⁵ Several elements were successfully pair matched and later confirmed with osteometric sorting.

The basic principle of osteometric sorting is that the two bones being considered are of a similar size and shape to have originated from the same individual. Osteometric sorting depends on the ability to distinguish anatomically normal size and shape relationships among skeletal elements. This is done by utilizing a reference sample to calculate means and standard deviations. The Forensic Anthropology Data Bank from the University of Tennessee, Knoxville was utilized because it consists of individuals of various age groups and ancestral origins. A database of non-standard measurements using the Osteology Skeletal Collection, curated at North Carolina State University, was created and utilized in this study.

Three models were used in the osteometric sorting process.⁵ Model one compares left and right sides, which accounts for shape differences, similar to visual pair matching and, in many cases, performs equally as well. It takes the form $D = \Sigma(a_i - b_i)$. The standard deviation of the difference between left and right sides in the reference sample was compared against the summation of differences from each pair of right and left elements and evaluated against a two-tailed *t*-distribution. A significant value (*p*-value <0.10) indicates that the two elements are significantly different and could not have originated from one individual. Model two compares articulation surfaces of bone elements based on the size of articulating surfaces. It takes the form $D = c_i - d_j$. In this model, the reference sample mean is subtracted from the difference between measurements of articulating bone portions and the total is divided by the reference sample standard deviation. This value

is compared to the two-tailed *t*-distribution ($\alpha = 0.10$). Model three compares bones of different sizes. The available measurements of a bone are summed and the natural logarithm of the sum is used in the regression model. The 90% prediction interval was used. If the sectioning point representing the two bones falls within the prediction interval, the null hypothesis that the bones were of a similar size to have originated from the same individual is rejected.

Using model one, comparing left and right humeri, out of 48 left and right pairs, 17 could be segregated. Using model two, comparing femora and os coxae, out of 42 left articulations, 8 could be segregated. Using model three, for the comparison of humeri and femora, the regression equation, $HUM = -0.083(FEM) + 6.281$, 5 pairs were evaluated. No significant difference was found between any of the pairs; segregation based on osteometric sorting is not possible in this case and further testing would need to be performed. The use of gross and osteometric techniques could provide a first step in segregating commingled individuals, which could eliminate the need to perform DNA profile testing on each individual bone.

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Commingling, Osteometric Sorting, Skeletal Remains

H15 Histology at the Central Identification Laboratory as a Method to Analyze Pathological Conditions in Modern Human Osseous Remains

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After attending this presentation, attendees will understand how histological techniques used at the Central Identification Laboratory (CIL) at the Joint POW/MIA Accounting Command (JPAC) were used to identify a pathological condition in human osseous remains and how these techniques could be used to distinguish inconclusive osseous remains from non-human osseous remains.

This presentation will impact the forensic science community by providing an example of how our normal histological analytical techniques were used to identify human osseous remains which could have been easily misinterpreted as non-human due to their pathological alterations that lead to abnormal anatomical morphology.

The CIL has consistently focused on the use of various types of validated methods for the assessment of osseous remains, including metric analysis, chest radiographic comparison, and

various types of DNA testing. Additionally, the CIL has incorporated histological analysis for the assessment of possible human osseous remains (inconclusive osseous remains) since 2006.

Numerous disease processes leave characteristic signatures on bone. Some of these processes are often recognizable at the macroscopic level, while others can be identified at the microscopic level. Whether gross or microscopic, hard tissue pathology can provide useful information for identification as well as contributing to the determination of cause and manner of death or the individual's health status at time of death. Bones that differ from their regular anatomical structure can easily be misinterpreted as non-human osseous remains. Histological analysis can aid in the identification of osseous remains with pathological conditions from non-human osseous remains and can additionally provide detailed information regarding the pathological processes differential diagnosis.

The utilization of common histological techniques as employed in modern forensic anthropological casework may significantly reduce the number of osseous remains misinterpreted as non-human osseous remains due to pathological changes.

An example is provided of a portion of a human fibula associated with forensic casework that displayed significant pathological changes. This fibula was sampled for mitochondrial DNA (mtDNA) analysis, as is common protocol at the JPAC-CIL. While the mtDNA analysis yielded a human signature, the gross morphological characteristics were not suggestive of human remains. A section of this fibular fragment was subjected to histological analysis and was identified as probable human due to histological characteristics of the pathological bone. The findings were indicative of a pathological process that led to fast-growing new bone formation inside the normal medullary cavity, which modified the cross-sectional morphology of the bone.

The methodology for standard histological analysis at the CIL utilizes three steps: embedding, sectioning, and analysis. Basic epoxy-embedding techniques are used to stabilize the fragment for thin sectioning. Then a thin-sectioning saw capable of making bone sections approximately 0.8mm thick is used and the specimen is cut transverse to its longitudinal axis. Finally, the thin section is attached to a glass slide, viewed under a standard light microscope at 50x magnification, and compared to our SOP decision matrix. This common methodology used for distinguishing human osseous remains from non-human osseous and non-osseous remains can also be used for analyzing possible pathological conditions and gathering additional information regarding individual health status at time of death. The example given in this presentation describes how histological analysis of osseous remains can distinguish osseous remains with pathological conditions from non-human osseous remains, which may avoid misinterpretations of the gross morphological characteristics.

Physical Anthropology, Histology, Pathological Conditions

H16 Thoracic and First Two Lumbar Vertebral Ring Epiphyseal Union in Skeletal Age Estimation: Testing a Revised Method of Documenting the Maturation Process

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After attending this presentation, attendees will gain an understanding of the use of a revised method of documenting stages of epiphyseal union for the thoracic and first two lumbar vertebral centra to better comprehend the pattern, sequence, and timing of maturation of this skeletal age indicator and its utility in

forensic casework.

This presentation will impact the forensic science community by introducing a revised method of estimating vertebral ring epiphyseal union for the determination of skeletal age-at-death of adolescents and young adults. When this method is used with other skeletal age indicators, it may improve the accuracy of age estimation in human identification.

The progress of epiphyseal union was documented for the annular rings (epiphyses or secondary ossification centers) of the vertebral centra, both superior and inferior, including the twelve thoracic vertebrae (T1-T12) and the first two lumbar vertebrae (L1-L2) when available, for an existing autopsy sample of 46 individuals (14 females, 32 males) ranging in age from 11 to 31 years at death, housed at the Human Osteology Laboratory at the University of North Carolina Wilmington.

The progress of epiphyseal union was noted in stages (stages 0-4) representing no union through completed union. The stages were as follows: Stage 0, ring absent, centrum unfused; Stage 1, partially fused ring, sections of centrum fused and unfused, presence of gaps; Stage 2, complete fusion, clear demarcation between ring and centrum, no gaps but there may be a groove, no remodeling; Stage 3, complete fusion, ring edges are melded with centrum leaving no clear demarcation in portions of the centrum, a slight groove may be seen in some areas but centrum is fully remodeled in other areas; and, Stage 4, complete fusion, ring edges are melded with centrum leaving no clear demarcation in portions of the centrum, bone is remodeled with no grooves. This modified scoring method included five stages of union and differs from the previously studied four-stage method in that a greater distinction was made between varying appearances of complete union, specifically those instances with complete union having a groove present and complete union without a groove.

Results of using this modified scoring method with five stages indicated that thoracic and first two lumbar vertebral ring union correlated rather well with age-at-death, $r = 0.81$, $p < 0.05$. This correlation was higher than previously reported findings for vertebral ring union and age-at-death using four stages to record vertebral ring maturation. Males showed a higher correlation ($r = 0.82$) when compared to females ($r = 0.77$); however, there was no statistically significant difference in vertebral ring union mean values between females and males. Raw data observations showed that females generally matured at earlier ages than males. Regarding the sequence of union, there was a tendency for epiphyses to unite either from cranially oriented and caudally-oriented epiphyses before those toward and in the middle of the vertebral column, or for more caudally oriented epiphyses to unite in a cranial direction.

Findings from this study may be considered in forensic cases of unknown identity, mainly for corroborating information from other skeletal age indicators, as multiple lines of evidence from various skeletal age indicators are preferred. Further, if only vertebrae are recovered in forensic cases, use of this method may provide a general age range at death.

Vertebral Maturation, Age Estimation, Epiphyseal Union

H17A Review of the Calce (2012) Method for Estimating Age-at-Death From the Acetabulum

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After attending this presentation, attendees will have a greater understanding of the human acetabulum and its use as an age indicator.

This presentation will impact the forensic science community by providing the results of further investigation into the

traits and statistical analysis for estimating age-at-death from the human acetabulum.

The goal of this presentation is to inform attendees on the performance of the recently developed Calce method for assessing age-at-death utilizing the acetabulum through a phase-based approach.¹

Accurate estimation of age-at-death is of important value to the forensic anthropology community as it provides information toward building a biological profile. While a number of methods can be employed for age-at-death estimation, continued development on the utility of less-researched regions of the human skeleton is of importance for expanding on our understanding of how age is physiologically represented in bone.

The Calce method approaches age-at-death estimation through use of the acetabulum by gross observation of three traits with the goal of defining three broad age ranges: young adult, 17-39; middle adult, 40-64; and, old adult, 65+ years-at-death.¹ The value of an accurate method for determining age over 65 is of great interest due to the current lack of methods that provide the ability to accurately and precisely distinguish these older individuals from those of middle-age adult (40-64).

The method is applied through combined observation of three traits that develop through age: (1) acetabular groove; (2) rim porosity; and (3) apex activity. Use of this method is not sex-specific and was developed on a modern sample drawn from the Grant Collection curated at the University of Toronto and the William M. Bass Donated Skeletal Collection (WMBDSC) located at the University of Tennessee.¹

The study sample consisted of a bilateral sample of 979 ossa coxae from 499 individuals drawn from the WMBDSC. The WMBDSC was chosen for its significance as the most comprehensive modern American sample so as to provide the most forensic benefit as a reference population. The sample includes male and female individuals of known age. The sample was evaluated blind to all demographic data. In this study, both left and right sides were observed to test differences in asymmetry. The order of evaluation was staggered to prevent introduction of bias in scoring.

Statistical analysis to determine correct age estimation was conducted on a bilateral sample of 247 ossa coxae representing 126 individuals. In this sample, the range of age includes 35-94 years-at-death, with an average of 63.5 years and a standard deviation of 13.4. Individuals were sorted into the correct age range 68.4% of the time. When evaluating for asymmetry in score assignment, the left and right side have an accuracy of 67.7% and 69.1%, respectively.

Of the 126 individuals, only 121 in this sample contained both os coxa in good condition. Average age of this subsample is 63.2 years with a standard deviation of 13.5. From this sample, it was found that symmetrical scores (the same score given to both the right and left os coxa of the same individual) were assigned 84.3% of the time.

To assess for precision, 224 ossa coxae representing 114 individuals were randomly selected for re-evaluation. Correlations between first and second observations resulted in an intra-observer error kappa test value of 0.79, representing substantial agreement between observations.

For inter-observer error in scoring, a peer with a similar level of training in skeletal biology utilized the same sample that was used in evaluating intra-observer error. The scores observed in this evaluation were compared against the original scores. Inter-observer precision was found to have a kappa value of 0.54, representing moderate agreement between scores assigned by observers.

Results of this data analysis suggest that the accuracy of this method, to be 68.4%, is lower than the reported 81% accuracy by Calce.¹ While intra-observer error has a substantial kappa

value of 0.79, the inter-observer error was found to be kappa = 0.54. These findings suggest that the Calce method for aging the acetabulum might benefit from a re-evaluation of the traits and descriptions used to assign age or that they require the user to be well-versed in the variability of the human acetabulum.

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Age Estimation, Acetabulum, Forensic Anthropology

H18 Analysis of Non-Metric Subadult Sex Determination Traits in Four Samples of Known Age and Sex: Sex Determinants or Population Variants?

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After attending this presentation, attendees will gain an understanding of the morphological variation of two non-metric traits in four different subadult skeletal samples previously determined to be sexually dimorphic.

This presentation will impact the forensic science community by: (1) demonstrating differences in two non-metric traits within each sample analyzed; (2) showing how these traits vary between four populations; and, (3) comparing these results to previous studies of subadult skeletal material. This presentation will add to the research on reliability and repeatability of forensic anthropological methods in sex determination through the analysis of subadult skeletal samples with known age and sex compared to previously published rates.

Finding a subadult sex determination method has long been the goal of biological anthropologists. For more than 100 years, dimorphism has been noted in the subadult pelvis.^{1,2} This is the most dimorphic area in the adult skeletal system and many studies have been tantalizingly close to finding distinctive traits in subadults.³⁻⁶ However, quantifying the dimorphism observed and creating reliable and repeatable techniques which meet *Daubert* requirements for sex determination in subadults have proven elusive.⁷ Part of the reason for a lack of standardization may relate to population variation, as most studies examined one population with few cross-population evaluations conducted. This is primarily due to the limited number of collections of juvenile material of known age and sex available for analysis.

This study examines four samples for the two nonmetric traits outlined in Byers' *Introduction to Forensic Anthropology* as potentially dimorphic in subadults, the auricular surface elevation and greater sciatic notch angle.⁸ For the auricular surface analysis, females exhibit a completely raised surface on all edges. Males demonstrate an auricular surface with most, if not all, of its edges, in the same plane as the ilium.^{4,6} The greater sciatic notch is evaluated as either "deep" for males or "shallow" for females.³⁻⁵

Samples examined for this study include the Forensic Fetal Osteological Collection (FFC) (n=113), a subadult component of the Hamann-Todd Collection (HTH) (n=37), and the Trotter Fetal Bone Collection (n=37), and the Scheuer Collection (n=15). Only individuals with unfused ilium were analyzed as fusion of the ischium at the acetabulum alters the shape of the sciatic notch.

This study could not establish a relationship between auricular surface elevation and biological sex. In the FFC sample, 72% of males and 39% of females were correctly identified. The HTH component had poorer results, with 57.6% of males and 30% of females correctly identified. The Trotter collection showed no

reliability in these features for determining sex; 12.9% of males and 9.5% of females were correctly identified. In the Scheuer collection, males were correctly identified at 80%, but female accuracy was 20%.

When the sciatic notch was examined, no consistent pattern was discerned. In the FFC sample, males were correctly identified 40% of the time, females 53%. In the HTH collection, a higher rate for males was determined (70%); however, female rates were much lower (27%). In the Trotter collection, 70% of males and 67% of females were correctly identified, the best of the four samples. Lastly, the Scheuer collection showed 100% accuracy for males but 0% for females. The small sample may have contributed to this unusual result.

While finding a method for subadult sex determination is a continued goal for biological anthropologists, the difficulty lies in translating the dimorphism observed into quantifiable and reliable methods. Therefore, multiple populations must be compared to ensure that traits found to be dimorphic are not specific to one group, but are found across populations. This study demonstrates that two nonmetric traits previously studied and shown to be dimorphic fail to exhibit the same results when compared to additional samples. Further research is needed to more fully understand these traits across populations.

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Subadult Sex Determination, Sciatic Notch, Auricular Surface

H19 Quantification of Sex and Population Differences in Obturator Foramen Shape

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After attending this presentation, attendees will understand how the shape of the obturator foramen varies across sex and population groups.

This presentation will impact the forensic science community by quantitatively testing classic descriptions of sex differences in obturator foramen shape, thereby providing accuracy

rates and an objective analysis of shape differences that can be utilized in future forensic methods.

The pelvis is known to be the most reliable skeletal element for sex estimation techniques. In particular, the pubic region has been shown to be highly diagnostic, with females displaying a relatively "stretched out" appearance compared to males (i.e., relatively longer and thinner pubic rami, broader pubic bodies, and greater subpubic concavity). Given that the obturator foramen is encompassed within the pubic bone, it is not surprising that traditional texts also describe sex differences in foramen shape. More specifically, females have been suggested to display a relatively more triangular obturator foramen shape compared to a more circular/ovoid appearance in males.¹ Despite the qualitative descriptions presented in texts and used by anthropologists in visual examinations, quantitative analyses statistically testing for sex differences in obturator foramen shape are generally lacking in the forensic literature. The aim of this study was to perform a geometric morphometric analysis on obturator foramen shape to quantitatively test for both sex and population differences and evaluate the reliability of this trait in forensic applications.

The sample consisted of 329 ossa coxae from adult females and males across five different population groups. These population groups included three modern samples (U.S. Whites, U.S. Blacks, and Portuguese) and two archaeological samples (medieval Nubian and plains Native Americans). Standardized photographs were taken of the medial surface of the obturator foramen with both pubic rami lying flat on a table surface. For each specimen, the outline of the obturator foramen was then extracted using Photoshop®, and an elliptical Fourier analysis and subsequent principal component analysis were performed using the SHAPE™ v1.3 software.² Multivariate Analysis of Variances (ANOVAs) were then conducted to test for sex and population differences in the shape components.

Results indicate that the majority of shape variation exhibited by the obturator foramen is in relative height (PC1, 42% of variation). PC2 (20%) reflects the position of the inferior angle of the foramen, while PC3 (10%) captures the traditional "triangular" versus "ovoid" shape changes. When samples were pooled, significant sex differences were revealed in all three components ($p < 0.001$). In general, females display a relatively taller and more triangular obturator foramen with a more medially placed inferior angle than males. These results support the written qualitative descriptions provided in traditional texts. When all PCs were analyzed in a Discriminant Function Analysis (DFA), a correct sex classification rate of 76% was obtained using a leave-one-out cross-validation method. Within-population accuracy rates ranged from 69-83%, cross-validated. Using only PC3, representing the "triangular" versus "ovoid" shape changes, accuracy rates dropped to 60%. Even within-population DFA results displayed low correct sex classification rates (54-66%). Significant population differences were also observed on PC1 and PC3. Overall, the archaeological samples displayed relatively taller obturator foramina than the modern samples ($p < 0.001$), and the U.S. Black sample displayed significantly more triangular foramina than all other groups ($p < 0.001$).

These results suggest that although there are significant sex differences in obturator foramen shape that are consistent with previous descriptions (females display more triangular obturator foramina than males), the accuracy rates for this particular shape trait alone are not acceptable for forensic analyses. In order to obtain moderately high classification rates, other aspects of the obturator foramen shape must be included in the analysis (e.g., relative foramen height). Furthermore, significant population differences were observed, indicating that any potential sex estimation methods using the obturator foramen should include population-specific standards.

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Sex Estimation, Obturator Foramen, Geometric Morphometric

H2O Effects of Age, Sex, and Height on Cortical Thickness of the Distal Femur: Ankara Specimen

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The goal of this presentation is to explore relationships of the thickness of the cortex of the distal femur of age, gender, and height in a cross-section of the Ankara population by Magnetic Resonance Imaging (MRI).

This presentation will impact the forensic science community by exploring dimorphism and information obtained using radiological methods which can help forensic research.

Criminals may destroy the remains of their victims to make their identification difficult.

Left and right knee MRI images from individuals without trauma-related fractures and mass lesions or deformities were collected between May 1, 2013, and July 25, 2013, at the Radiology Clinic of Ankara Numune Hospital. T1-weighted images were studied to take advantage of hyperintense signal in the bone marrow fat. On midcoronal section, medial and lateral cortical thickness measurements were obtained (8cm proximal of the median level of femoral lateral condyle.) On midsagittal section, anterior and posterior cortical thickness measurements were obtained (8cm proximal of the median level of femoral lateral condyle).

The group consisted of 68 subjects (34 male and 34 female). The mean age was 45.80 years (18-81 years), the mean body height was 169cm (150cm-200cm), the mean weight was 79.26kg (52kg-120kg), and the mean Body Mass Index (BMI) was 27.73 (18.20-39.06).

In terms of gender with right medial and posterior cortical thickness, about 67.6% of the subjects were correctly classified. From the four quadrants, strong correlations were observed in the right medial femoral cortical thickness with age ($r=0.349$; $p=0.004$) and thickness of the cortex of the left medial femoral with height ($r=-0.337$; $p=0.005$).

By digital radiological studies in with Maximum Vertical Diameter (VHD), Harma and Karakaş could correctly classify 77% of the Anatolian Caucasians and Mell and co-workers could classify 86.8% of the German subjects.^{1,2} Literature on the femoral cortex regarding the relationship between age, sex, and height tries to explain the population variation.³⁻⁵ Together, the existence of such dimorphism and information obtained using radiological methods can aid forensic research.

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Cortical Bone, Magnetic Resonance Imaging, Forensic Anthropology

H21 A Preliminary Assessment of the Effects of Observer Error in the Consistency of Age-at-Death Estimates Based on the Pubic Symphysis

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After attending this presentation, attendees will identify some of the difficulties in applying visual scoring methods for adult age-at-death estimation, based on morphological changes of the pubic symphysis, and evaluate their implications for the precision and accuracy of age estimates.

This presentation will impact the forensic science community by demonstrating how different adult age estimates can be in any given forensic case, depending on who is scoring and estimating age.

Apart from the estimation of sex, stature, and ancestry, the estimation of age-at-death is one of the most important parameters in the early stages of forensic identification, during the reconstruction of the biological profile. Numerous methods have been developed for adult age-at-death estimation from skeletonized and badly preserved human remains, most of them based on visual scoring of morphological indicators of age, such as those of the pubic symphysis. The purpose of every method is to be reliable, accurate, and easy to use by different researchers and using distinct samples. However, due to the complexity and variation in the aging processes, age estimation is not always as precise as required. The purpose of this study is to test how consistently four different investigators assign age-at-death to five adult skeletons based on the morphological features of the pubic symphysis. The skeletons (three females and two males) were selected from the Human Identified Skeletal Collection that is being amassed under the BoneMedLeg Project (<http://bonemedleg.host22.com/>) in Porto, Portugal, and age was estimated using the method proposed by Milner and Boldsen, where five separated components were considered: the symphyseal relief; the dorsal symphyseal texture; the superior protuberance; the morphology of the ventral symphyseal margin; and, the dorsal symphyseal margin.¹ The score for each component was then entered in the ADBOU age estimation software developed by Boldsen and co-authors (2002) to estimate the maximum likelihood age and its respective 95%

confidence interval.² Two measures of precision of this method were calculated: (1) estimation of inter-observer agreement rate for each morphological component; and, (2) inspection of inter-observer agreement for the 95% confidence interval of age obtained from the software. A low rate of inter-observer agreement — ranging between 52% and 77% — was obtained. Comparison of 95% confidence intervals of age show significant discrepancies between different observers, but these differences are only more noticeable at the lower or upper end of the confidence interval. These discrepancies in the 95% confidence intervals of age are partly due to variation in inter-observer error and also to factors not completely identified but perhaps related to a lack of significant association of some of Milner's and Boldsen's method components with age. When such an association is low or nonexistent estimates of age obtained from different observers will be equal or similar even when there is little or no inter-observer agreement for the stages in the different components of the method.

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Age Estimation, Pubic Symphysis, Interobserver Error

H22 Differences in Skeletal Pathology as Seen in an Individual With Quadriplegia Secondary to Duchenne Muscular Dystrophy

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After attending this presentation, attendees will see the distinction between a human adult skeleton displaying the effects of Duchenne Muscular Dystrophy (DMD) with normal adult skeletons. This will aid anthropologists in differentiating between normal bone variation and bone pathology. Although the skeleton is similar in size to a subadult or young female skeleton, the systemic loss of bone mass and density of the postcranial skeleton will reveal that this is not normal human variation but a pathological condition.

This presentation will impact the forensic science community by bringing awareness of what effect Duchenne Muscular Dystrophy has on the skeleton and how this may be distinguished from other bone disorders such as osteoporosis or normal bone variation.

The Mechanostat Model describes the direct correlation between muscle use and bone strength and density. If the muscle is active, applying force to bone with a strain greater than 1500 μ , the bone will respond by increasing in strength and mass. Bones of individuals who have sustained some level of paralysis, and thus strain less than 800 μ , will have a reduction in strength and mass.

In this study, the body of a deceased 63-year-old White male was donated to the Southeast Texas Applied Forensic Science (STAFS) facility with quadriplegia secondary to DMD, was compared to the bones of four White males of similar age with no paralysis history. Comparisons included the examination of the gross morphology, three-dimensional morphometry,

histomorphometry, and bone density analysis. Three-dimensional data was collected using the Microscribe® G2X and analysis was completed using Morpheus *et al.* software. Bone histomorphometry examined osteon size, osteon number, osteon area, and osteon density. Dual Energy X-Ray Absorptiometry (DEXA) was used to determine Bone Mineral Density (BMD). Postcranial measurements were also taken using conventional methods (i.e., sliding caliper, osteometric board).

Except for the cranium, the gross morphology of the DMD skeleton showed systemic reduction in bone mass and density with the overall size similar to that of a young adult or a female. The skeleton was gracile with small articulating surfaces (e.g., femoral and humeral head sizes).

Results for cranial morphometrics show that the majority of the mean measurements were statistically significant ($p < 0.05$) between the DMD subject and the four normal subjects. With the exception of MDH, OBH, FOL, OCF, CDL, and MAN, all other cranial measurements were substantially larger for DMD than the other subjects.

All postcranial mean measurements between DMD and normal subjects showed statistical significance ($p < 0.05$) with substantially larger postcranial measurements on the four normal subjects.

Studies using osteon size, area, and density to estimate age show osteon area for individuals older than 50 years of age typically range between $12,868\mu\text{m}^2$ - $37,762\mu\text{m}^2$ (Crowder 2012). Osteon diameter of bones with disuse, osteoporosis, or age-related osteoporosis is larger and the density lower (Black 1974). The study subject (DMD) had fewer osteons ($n=3$), larger osteon diameter ($317\mu\text{m}$) and area ($78,923\mu\text{m}^2$), and a lower concentration of osteon population density ($1.46/\text{mm}^2$).

Previous research on White males from the United States show the average BMD is approximately $0.790\text{g}/\text{cm}^2$ (Looker 1995). Average BMD T-scores for females in the United States ranges between -1 and 1. The Dual-Energy X-Ray Absorptiometry (DEXA) scan for the DMD male showed a BMD of $0.466\text{g}/\text{cm}^2$ and a T-score -4.2. The T-score of the DMD male is significantly lower than found in his ancestry and age range and significantly lower than the average female. Conversely, one of the normal male subject's BMD score was $1.088\text{g}/\text{cm}^2$ and T-score 2.1 showing a slightly higher density than the average.

Results of the Pearson's correlation (to determine if there was a relationship between cranial and postcranial measurement of DMD) showed no dependent relationship. This lack of correlation underscores the limitations of the Mechanostat Model in that it does not adequately explain the difference in bone biomechanics and remodeling between the skull (non-weight-bearing) and postcranial bones (weight-bearing).

Bone Pathology, Duchenne Muscular Dystrophy, Mechanostat Model

H23 Metric Assessment of Sexual Dimorphism in the Scapula: A Validation Study

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After attending this presentation, attendees will understand the utility of the scapula in forensic sex estimation in adults.

This presentation will impact the forensic science community by widening the array of validated methods available for sex estimation of skeletal individuals, thus complying with evidentiary standards outlined in the *Daubert* guidelines.

Furthermore, variety in methodology involving single

skeletal elements increases the accuracy of the biological profile constructed for individuals with missing or damaged elements that are commonly referenced, especially when used in conjunction with other single-element methods in a multifactorial approach.^{1,2}

The purpose of this study is to test the validity of a two-variable method of metric sex estimation using the maximum breadth and maximum length of the scapula, as introduced by Dabbs and Moore-Jansen in 2010.³ In the Dabbs and Moore-Jansen study, two discriminant function equations are presented, one involving two variables and one involving five variables. The two-variable function has greater applicability to researchers due to the simplicity of the measurements and instruments required, and therefore is the subject of this validation study. The scapular breadth and length measurements are inputted into this discriminant function equation and the result is a value greater or less than zero, the male-female threshold. A positive value classifies the scapula as male, and a negative value classifies the scapula as female. This method was developed on late 19th-early 20th-century Black and White individuals from the Hamann-Todd collection. However, it has been shown that individuals from this era do not fully represent the variation seen in modern populations.^{4,5}

This retrospective study tests the Dabbs and Moore-Jansen two-variable method on 73 scapulae from a total of 38 positively identified decedents brought to the Harris County Institute of Forensic Sciences (HCIFS) for autopsy pursuant to Article 49.25 of the Texas Code of Criminal Procedures. At autopsy, these decedents were unidentified and in varying stages of decomposition, from moderate to advanced. Therefore, anthropological examinations were required to assist in identification or other supporting analyses.

For this study, all available complete scapulae were analyzed to first assess effects of bilateral asymmetry. This sample included a total of 73 scapulae from 38 females and males of Black (F=2, M=7), White (F=8, M=16), and Hispanic (F=1, M=4) ancestry. All were represented by bilateral scapulae in the sample except for one White male, one White female, and one Hispanic male, who were represented unilaterally. The Hispanic sample was evaluated to test the method's applicability on non-Black and non-White populations. Anthropological case reports and bench notes written by four different HCIFS anthropologists were reviewed and the maximum scapular breadth and maximum scapular length measurements were recorded. These measurements were then entered into the Dabbs and Moore-Jansen discriminant function to estimate sex, and the results were compared to the decedent's known sex. Bilateral asymmetry had no effect on the classification of any individuals. Thus, the left scapula (if unavailable, the right scapula) was selected from each of the 38 decedents to calculate accuracy in percentage of correct sex classifications.

Results suggest that performance of the two-variable method is best when applied to certain subgroups within forensically modern populations. When using the method on Black and White individuals, the overall accuracy is 93.75%. Within the Black population, the accuracy is 77.78% due to misclassification of both Black females. The two-variable method performed very well within the relatively large White population with 100% correct sex classification. In Hispanic populations, accuracy is 60% due to misclassification of two males as female. As noted by Spradley *et al.* there is a tendency for misclassification of Hispanic males as female in metric analysis.⁶ An overall accuracy of 89.47% is observed when applied across all populations.

This study demonstrates that the Dabbs and Moore-Jansen two-variable method for sex estimation using scapular metrics is potentially a useful tool for forensic anthropologists when analyzing unknown skeletal remains. However, the method presents low accuracy when applied to the small samples of HCIFS Black females and Hispanic males, suggesting the need for a larger test sample with stronger representation of each subgroup

to determine true applicability.

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Sexual Dimorphism, Metric Analysis, Scapula

H24 The Use of Enthesopathies in the Femur and Os Coxa for Assessing Age

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After attending this presentation, attendees will understand the value of enthesopathies in the femur and os coxa for assessing age.

This presentation will impact the forensic science community by examining a new method of age prediction from skeletal remains.

Enthesophytes commonly are examined in bioarchaeological research for their relationship to activity. While the value of these markers for assessing activity patterns remains questionable, results from such studies consistently show that they are significantly correlated with age in historic and prehistoric populations. The present study explores whether enthesophytes in the femur and os coxa are correlated with age in a contemporary population and, if so, can be used to: (1) generate age predictions, and, (2) narrow age estimates for older individuals.

The study sample is comprised of 200 White individuals (100 of each sex) over 40 years of age from the William M. Bass Donated Collection. Four areas on the femur and five on the os coxa were scored for the degree of enthesophyte formation based on methods modified from Mariotti and colleagues.¹ These areas included the lesser trochanter, spiral line, gluteal tuberosity, linea aspera, iliac tubercle, posterior iliac crest, ischiopubic ramus, and inferior and posterior ischium. Using raw scores, intra-observer error was assessed in a subsample of 30 individuals using the Wilcoxon signed rank test. For the entire sample, chi-square was used to examine the relationship between raw scores and ten-year age categories. Additionally, because several studies note a correlation between body size and enthesophyte size, raw scores were adjusted for body size by dividing each score by femoral head diameter. Linear regression analyses then were used to examine the relationship between adjusted scores and age.

Results indicate the following. Significant intra-observer differences were noted in one variable, posterior iliac crest ($Z=-2.121$, $p=.034$); therefore, this variable was excluded from further

analyses. Based on raw scores, among males, five of eight variables (lesser trochanter, gluteal tuberosity, linea aspera, ischiopubic ramus, and posterior ischium) are significantly correlated with age category. In females, only three of eight (spiral line, iliac tubercle, and posterior ischium) are significantly correlated.

Individual regression analyses show that adjusted scores for all variables are significantly correlated with age in males; however, in females, only four are significantly correlated (lesser trochanter, iliac tubercle, and inferior and posterior ischium). None of the variables exhibits particularly strong associations (R^2 ranges from 0.039 (posterior ischium, females) to 0.176 (lesser trochanter, males)). Forward Stepwise regression indicates that, for males, the lesser trochanter ($\beta=233.171$, $p=0.002$) and gluteal tuberosity ($\beta=219.6244$, $p=0.005$) are the crucial predictors of age ($F=14.466$, $p=0.000$, adjusted $R^2=.226$). For females, the lesser trochanter ($\beta=276.11$, $p=0.001$), linea aspera ($\beta=-243.01$, $p=0.013$), and iliac tubercle ($\beta=241.00$, $p=0.000$) are the crucial predictors ($F=11.697$, $p=0.000$, adjusted $R^2=0.245$).

Lastly, the frequency of the most severe enthesophyte expression ("Score 3" or "gross morphological alteration") was examined to determine its use for narrowing age estimates in older individuals. Among males, for four variables (lesser trochanter, spiral line, gluteal tuberosity, and ischiopubic ramus), 75% of individuals with the most severe expression were aged 70 years and above. In fact, for the latter three variables, the severe expression was never found below 60 years. Among females, for six variables (lesser trochanter, iliac tubercle, inferior ischium, spiral line, gluteal tuberosity, and linea aspera), 70% of individuals with the more severe expression were aged 70 years and above; for the latter three variables, the severe expression was never found below 60 years.

The current study demonstrates that significant relationships exist between age and enthesophyte scores in the femur and os coxa, but the relationships are not strong enough to generate reliable age predictions. Confounding factors might include methodological limitations: neither ordinal categories nor simple visual assessment appears to be sufficient for capturing enthesal morphological variation within age categories. Conversely, the frequencies and age distribution of scores suggest they can be used to corroborate age assessments based on other methods. Additionally, in situations where traditional areas used for aging are not available, gross morphological alterations at certain entheses can suggest an age greater than 60 years or, in some cases, 70 years.

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Age Estimation, Enthesopathies, Anthropology

H25 Forensic Anthropology Education and Mentoring Opportunities at the University of Tennessee Forensic Anthropology Center

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After attending this presentation, attendees will understand some of the parameters of the proposed "Specialist certification level" of the American Board of Forensic Anthropology (ABFA) and the new training initiatives the University of Tennessee Forensic Anthropology Center is doing to prepare students for Specialist certification.

This presentation will impact the forensic science community by providing one model for advanced training of students in academic forensic science disciplines that explicitly utilizes current laboratory accreditation and professional certification standards as guidelines for forensic science mentoring.

The purpose of this presentation is to demonstrate how innovative training and mentoring initiatives of the Forensic Anthropology Center (FAC) at the University of Tennessee, Knoxville, are preparing undergraduate and graduate students for careers in forensic anthropology. Importantly, these training efforts are geared to prepare students for the proposed American Board of Forensic Anthropology (ABFA) certification changes.

For over 35 years, ABFA has offered Diplomate certification to post-PhD forensic anthropology professionals. Very soon a new ABFA "Specialist" certification opportunity will be available that targets post-MA students and professionals. Proper preparation for certification will require a shift in the pedagogical framework of academic-based disciplines. While coursework should not be de-emphasized, academic institutions must provide a variety of experiences that provide both students and junior colleagues with the requisite specialized training and casework to achieve certification.

The FAC has recently implemented a number of initiatives that drive the Center toward laboratory accreditation as well as Specialist and Diplomate certification. There are three basic mentoring goals for students: (1) provide professionalization training that builds a minimum skill set to achieve certification-level standards in the field; (2) provide access to cases and formal case reviews with mentors to ensure best practices are employed and to teach technical writing skills necessary for clear and concise case reports; and, (3) provide both academic as well as social networks that enable close mentor-student interactions outside of the classroom. To prepare for Diplomate certification, the FAC has established a formal rotating peer-review system in which one of the six forensic anthropology professionals serves as the case lead while the others peer review the case. Graduate students are also involved in casework on a rotating basis.

The FAC achieves the mentoring goals in a number of ways. Professional training courses are available on a quarterly basis to all Anthropology graduate students. This typically involves day-long trainings on specific subjects, such as standard laboratory procedures, equipment-based training (e.g., total station, GIS, 3D digitizer), proper archaeological excavation techniques, and appropriate usage of computer software and statistics. Each session is explicitly framed by pertinent theoretical, ethical, and professional discussions. Participants receive certificates of course completion and the FAC director documents the training hours for each participant to log for certification preparation.

The key to success of these initiatives is to provide multiple opportunities for students to apply the skill sets established by their coursework and the professional training sessions. For instance, all participants who attended the Body Donation Program training in Spring 2013 are eligible to assist FAC staff with daily tasks at the FAC, including donation intake, biological sampling, laboratory work, and body placement at the Anthropology Research Facility (ARF). Further, all participants are eligible to conduct formal casework with the faculty mentors. Students keep a log of their case-related activities that is signed by a Diplomate.

In addition to participation in formal forensic casework, the 1,100 skeletons of the Bass Donated Skeletal Collection can serve as proxies for case studies. While all UTK students are eligible

to study the skeletons, a formal system has been established to encourage casework participation and close mentoring by the FAC faculty. Two donated skeletons are selected bimonthly by the FAC Director for student examination. Provided with a scenario and problem, students conduct an analysis and submit a written report and bench notes to one or more faculty mentors, including a board certified forensic anthropologist. The strengths, weaknesses, and accuracy of the analyses and reports are discussed with each student.

In conclusion, the FAC uses laboratory accreditation and professional certification as pedagogical roadmaps for forensic science mentoring. One ultimate measure of success of the training initiatives will be determined by the number of successful Diplomate and Specialist certifications obtained by students and faculty.

Mentoring, Forensic Anthropology, Certification

H26 The Development of Professional Standards and Accreditation by the British Association for Forensic Anthropology (BAFA)

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After attending this presentation, attendees will understand the rationale behind the development of the United Kingdom accreditation system for forensic anthropologists and have an understanding of how the process works, how it complements the requirements of the criminal justice system, and how it supports the professionalization of the discipline.

This presentation will impact the forensic science community by communicating the development and rationale of the accreditation system for forensic anthropologists in the United Kingdom which has been developed to support the professionalization of the discipline, provide a career pathway for future forensic anthropology practitioners, and ensure that end-users understand the skill levels of the professional they are employing to undertake casework.

Accreditation of forensic practitioners in the United Kingdom has gained momentum in recent years and has been driven by a series of high-profile miscarriages of justice. While large forensic science service providers have established means by which they can demonstrate and maintain accreditation, this has proven more difficult to achieve for the independent practitioner, including the practicing forensic anthropologist.

As a result, the British Association for Forensic Anthropologists was formed and has undergone an intensive period of standard setting and validation under the auspices of our professional body, the Royal Anthropological Institute of Great Britain and Ireland (RAI). This has included the development of a Code of Practice, Ethics, and Professional Standards, establishing Forensic Anthropology Practitioner Guidelines and a Continuing Professional Development and examination process based on a robust curriculum. These documents have been written by forensic anthropologists who are currently undertaking and presenting their case-work before the United Kingdom judiciary.

This presentation will describe the development and rationale of the practitioner accreditation process which has now been fully ratified by the RAI and put into practice. The BAFA

accreditation process aims to provide two things; a transparent, fit-for-purpose guide to end-users on the skills and experience of the forensic anthropologist that they are employing and a definitive career pathway for the forensic anthropology practitioners of the future. The accreditation process and its development will be discussed within the framework of appropriate scientific quality standards now being demanded by the Forensic Science Regulator and the criminal justice system in the United Kingdom.

Accreditation, Forensic Anthropology, BAFA

H27 An “Insider” Look at Forensic Anthropology: Theoretical Grounding, Logical Reasoning, and Scientific Explanation

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The goal of this presentation is to explore the epistemological foundations of forensic anthropology. A focus is the applicability, accuracy, and limitations of three major forms of reasoning and argumentation utilized in forensic anthropology, which contribute to its status as a distinct, scientific discipline.

This presentation will impact the forensic science community by helping attendees to gain a clearer understanding of the theoretical grounding which defines forensic anthropology as a discipline. Meeting attendees will gain a clearer perspective on the differences between the major forms of logical reasoning, their effect on hypothesis generation and testing as well as evidence collection and interpretation, and their importance in the fields of scientific and legal argumentation.

A recent “outsider” view of forensic anthropology has questioned its legitimacy as a discipline and as a science due to the absence of a grounding body of theory. Forensic anthropology is seen as focused on applied “lab-based” activities; field recovery and documentation utilize established archaeological methods, but no broader theoretical development in the field is evident.¹

Scientific theory provides testable explanations for observed phenomena.² It is shown here that forensic anthropology routinely constructs and utilizes scientific theory, both in the lab and field. In arriving at explanations for observed incidences of trauma, taphonomic effects, and even the characterization of the biological profile, forensic anthropologists develop and utilize a broader theoretical base (including well-established forms of logical reasoning) than is suggested by the above criticism.

Construction of theory is dependent upon logical reasoning and argumentation. Three major forms of logical reasoning—deductive, inductive, and abductive argumentation—are used by forensic anthropologists to offer plausible explanations for the specific types of data they examine.³ As explanations of observable phenomena, they form the scientific theoretical foundation of forensic anthropology. In this presentation, each of these three forms of logical argumentation are defined, compared, and illustrated by examples from forensic anthropological research.

Deductive reasoning is based on the concept that if the premises of a statement are true, then the conclusion must also be true. Inductive inference is based on the statistical evaluation of a set of data and assessment of the probability that an explanation is correct. Although both inductive and deductive reasoning are components of the traditional model of scientific reasoning referenced in the *Daubert* standard, less rigorous or definitive abductive arguments are also a component in the development

of scientific knowledge. This form of reasoning is also called “inference to the best explanation” - the explanation that is most compatible with the available data.⁴ Abductive reasoning is context-dependent and most effective in providing explanations for rare or unique events (common to forensic anthropology), with the ability to incorporate large numbers of variables into an explanation. It is dynamic and falsifiable in that new data can invalidate initial explanations. Although abductive reasoning is the initial step in the development of scientific hypotheses, abductively-based explanations in themselves are critical for guiding evidence collection and interpretation. In forensic anthropology, abductive reasoning forms the basis for the majority of taphonomic studies, reconstructions of forensic scenes and events, and interpretation of trauma. Abductive arguments should not be excluded from scientific theory building (or courtroom testimony) for they are often compatible with both scientific and legal dialogue – the *Kumho* ruling allows the judicial system some flexibility in admissibility of arguments like these that have not been fully tested.⁵

Thus, much of what forensic anthropologists do is indeed scientifically rigorous, although not in the strict deductive-nomothetic sense in which science is often portrayed. There is a solid, albeit nascent, grounding body of theory (and theory building) inherent in forensic anthropology. The characterization of our discipline as atheoretical is not a “fatal flaw” – this depiction is both inaccurate and untenable.

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Forensic Anthropology, Theoretical Reasoning, Scientific Explanation

H28 Sex and Ancestry Determination: Assessing the “Gestalt”

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After attending this presentation, attendees will gain an understanding of assessment of sex and ancestral affinity from human crania and how previous training and experience can affect one’s ability to quickly and accurately assess sex and ancestry in a quiz-like setting.

This presentation will impact the forensic science community by providing information regarding how the assessment of sex and ancestry from the skull is affected by a combination of training and individual assessment of non-metric traits of the skull.

Visual assessment of morphoscopic traits of the human skull for sex and ancestry determinations is well-known in the anthropologists’ tool kit. Assessment techniques are taught throughout basic and advanced osteology classes and are heavily

relied upon in forensic case work. Multiple authors have examined the accuracy rates of scoring techniques in the literature. However, the extent to which these techniques are actually derived from an overall impression of the skull that is created within the first few moments of seeing it has not been assessed. The accuracy of the investigator's "gestalt" is explored through a short, timed test and then these results are compared to published accuracy rates.

A 20-skull test was offered during the DOD JPAC-CIL/ Missing Person Cases Symposium held last year. Fifty participants agreed to undertake the experiment (n=2,000). Each observer was given 20 seconds per skull to record the sex (male or female) and ancestry (White, Black, or Asian) for each specimen. Participants were asked to circle any answers that they wished to spend more time on. At the conclusion of the test, participants were allowed to re-examine circled answers for as long as they wished, and key an alternate answer, if so desired.

Prior to this test, this study's completed similar tests. One hundred skulls were blindly selected from the Smithsonian Institution's Terry Collection. The test was set up in a series of 5 sets of 20 (one was split into 10s) with 20 seconds allotted to assess sex and ancestry (White or Black) for each skull. Re-examination was allowed at the end of the test (for 50 skulls). At the conclusion of these tests, Dr. David Hunt selected another 20 skulls that contained three ancestral categories (White, Black, and Asian) from the holdings and the test was conducted again (total n=480).

For the first 100 skulls, sex was accurately determined in 82% and 84% of the cases, and ancestry was correctly determined in 85% and 89% of the cases, per investigator. When given the opportunity to re-evaluate the skulls for a longer period of time, the investigators frequently changed a correct answer to an incorrect answer. That is, they were more accurate when their first answer was unchanged. With the addition of a third ancestral category, sex was accurately determined in 85% and 80% of cases, respectively, and ancestry declined to 75% and 60%, respectively.

Results from the larger experiment indicated that sex and ancestry determination is linked to experience and highest degree achieved, with PhD holders out-performing those with lower degrees and/or degrees in related fields. Sex determination was accurately estimated 74% of the time for PhD's, followed by MA's (70%), BA's (69%), and alternate degrees (64%). Ancestry estimation followed a similar pattern, 68%, 55%, 57%, and 52%. A much larger gap is noted in ancestry estimation between degrees achieved, possibly due to the more complex characteristics used in this determination.

The study sex assessments are remarkably accurate, fitting well with published accuracy rates of morphoscopic traits, given the little amount of time spent on the decision. This suggests that each participant has their own "gestalt" about cranial features and that decisions are likely made in the first few seconds of viewing a skull. Similarly, the accuracy of ancestry determinations is relatively high, though not quite as high as several published examples. This suggests that morphoscopic traits are "checked-off" very quickly when examining a skull to provide a "gestalt" assessment. Arguably, much of the "gestalt" is due to initial training, followed by years of experience. A more detailed discussion of these results will be presented at the meeting.

Physical Anthropology, Sex, Ancestry

H29 Experimental Vertebrate Taphonomy: An Experiential Active-Learning Experiment Design and Implementation Course for Forensic Physical Anthropology Education and Mentorship

Thomas Evans, MA, MT State University, Dept of Earth Sciences, PO Box 173480, Bozeman, MT 59717-3480*

The goals of this presentation are to describe an undergraduate course in experiment design and implementation and inform as to the impact the course had on the students and instructor.

This presentation will impact the forensic science community by informing how course design can improve the content knowledge received by undergraduate forensic physical anthropologists about experiment design, as well as improve students' cognitive development so they are prepared to perform and implement conceptually more robust and useful experiments in their graduate careers.

Experiential active learning courses have the potential of cementing subject-specific content knowledge through solving authentic problems and developing student cognition by confronting students with achievable problems. An Experimental Vertebrate Taphonomy course is one method to teach experiment design concepts and stimulate transformative cognitive development by having students implement their own experiment designs.

Course Structure: The course is designed with two components; a discussion section three times a week for a total of three hours, and a laboratory section twice a week for a total of four hours. Discussion sections for the first half of the semester cover scientific behavior (e.g., ethics), experimental and historical science philosophy, and experiment design concepts. Laboratory section time is used to teach essential scientific skills such as how to perform literature searches, write and read scientific papers, write abstracts, make and give scientific talks, etc. To end the first half of the semester, the instructor designs an experiment to be performed by the entire class. The second half of the semester, students use discussion section time to write and edit papers or prepare and practice scientific talks. In the laboratory section, students implement experiments that they designed themselves.

Assessments: Students are graded on a take-home midterm and final, as well as an in-class midterm and final. The majority of course points are awarded for a submission-quality publication and a 15-minute oral scientific presentation. In addition, students are graded on a detailed written description of their proposed experiment that demonstrates their mastery of experiment design concepts, as well as a laboratory notebook that meets the professional standards of the National Institute of Health.

Samples: One of the greatest challenges of performing experimental taphonomic research is procuring samples in an ethical way that also provides researchers with statistically significant numbers of samples. Most universities have an ample supply of deceased laboratory mice, rats, or rabbits that are used for biological or biomedical research. If an animal resource center exists on campus they can be contacted and their dead can be procured for course use. During hunting season additional remains can be obtained from local meat processors, and juvenile remains can often be acquired from large farms during calving or lambing seasons. Lastly, fish remains can be either procured through legal fishing or from fish markets, and sometimes free heads are available after preparation for sale.

Qualitative Results: Students responded enthusiastically to the course, expressing sincere appreciation for the effort to engage them in authentic scientific research that could benefit

their careers. Many students commented that the course was the most transformative they had experienced in college, since they were expected to perform science and treated like fellow scientists rather than “just undergrads.” From the instructor’s perspective, it was clear that students grew as independent scientists and in their capacity for future scientific endeavors. They all grew in their capabilities of developing and implementing experimental taphonomic research and three students expressed a desire to further their taphonomic research at the graduate level.

Quantitative Results: Four students presented their work orally at a student research symposium at Montana State University, giving them valuable presentation experience. In addition, one student continued her research and is presenting her work here at the AAFS annual meeting this year.

Conclusions: The course was a resounding success and accomplished five objectives: (1) students learned how to design and implement rigorous taphonomic experiments; (2) students performed authentic research which was shared with the greater scientific community; (3) students had a transformative learning experience that stimulated a long-term interest in (taphonomic) experimentation; (4) students and instructors alike were invigorated, making research and teaching fun and exciting in ways not anticipated before the course; and, (5) lastly, lasting friendships were developed and fostered in a supportive learning environment.

Experiment Design, Taphonomy, Teaching

H30 The Evolution of Education and Mentorship: A Symposium in Honor of Norman J. Sauer

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After attending this presentation, attendees will have an understanding of the impact of successful mentorship from the undergraduate through post-doctoral level by looking at the Michigan State University program under the direction of Norman J. Sauer.

This presentation will impact the forensic science community by highlighting the evolution of forensic science education in a forensic anthropology program. It will discuss the far-reaching impact of a mentor in the field and address areas for future consideration in further strengthening the balance between forensic science education and professional mentorship.

The current physical anthropology program at Michigan State University began under T.W. Phenice in 1968, with an emphasis on making skeletal biology accessible to students of osteology. With the addition of Norm Sauer to the faculty in 1974, the program’s focus on education in skeletal biology and the application of the field to forensics was solidified. Sauer’s influence on the field extends past his contributions in research and publications to his devotion to the classroom. Whether through undergraduate curricula, graduate student mentorship, community outreach, professional law enforcement development, or leadership within the American Academy of Forensic Sciences, Sauer has exemplified what it means to be a mentor to future generations of forensic scientists.

This symposium is organized to examine which components are part of a successful mentorship program and highlights the changing roles associated with mentoring to multiple levels of students and professionals. The presentations included are examples of professionals impacted by Dr. Sauer’s efforts

and the avenues of education and research that have developed as part of that influence. They highlight the breadth of research interests initiated through an educational program, the institutions that now employ past students, and growing impact on both original research in the field and the expansion of educational programs in the United States.

In the following symposium, Wankmiller and Bird discuss the importance of graduate education on professional development through student-mentor relationships. They include a discussion of current research in the areas of facial image analysis, trauma analysis, and human rights. Kolpan and McCormick offer a look at the use of craniometrics and contextual information in assessing ancestry in unknown samples. Their research provides an example of professional research influenced by the research interests of undergraduate and graduate mentors. The presentations combine areas of bioarchaeological instruction with its application to forensic anthropology in practice.

The symposium offers the opportunity to discuss educational initiatives within the field of forensic anthropology and the importance of curriculum development and mentorship opportunities. As highlighted by organizations such as the Scientific Working Group For Forensic Anthropology (SWGANTH), National Science Foundation, and National Academy of Sciences, education and training are a vital part of the growth and development in the field of forensic science.

Mentorship, Forensic Anthropology, Education

H31 Athena in Disguise: The Mentorship of Dr. Norman J. Sauer During the Last Ten Years at Michigan State University

Jane Wankmiller, MS, Michigan State University, Dept of Anthropology, 355 Baker Hall, East Lansing, MI 48824; and Cate E. Bird, PhD, 4714 E Halifax, Mesa, AZ 85205*

After attending this presentation, attendees will have an understanding of the breadth of successful mentorship that Dr. Norman J. Sauer has demonstrated during his last ten years of tenure at Michigan State University.

This presentation will impact the forensic science community by assessing the mentor-protégé relationship between Dr. Sauer and two of his graduate students and his influence on both the direction of their research and their development as professionals.

One of the earliest documented accounts of mentorship comes from Homer’s *The Odyssey*. In this classic tale, Odysseus’s son, Telemachus, is aided by a character named Mentor, who happens to be Athena in disguise. This Mentor archetype has persisted today and remains a critical aspect of professional development in graduate education. Successful mentors serve as teachers, motivators, role models, advocates, and voices of reason and conscience. Dr. Sauer’s relationship with his protégés has not only been educational, but also profoundly personal and mutually respectful. Dr. Sauer’s breadth of expertise is largely confirmed by graduate research he supervised during the last decade at Michigan State University.

Jane Wankmiller has been Dr. Sauer’s student since 2005. Dr. Sauer’s breadth of expertise and mentorship is highlighted by projects on which he and Ms. Wankmiller have collaborated, involving diverse forensic cases and bioarchaeological research. In addition to traditional cases involving skeletal analysis, Dr. Sauer and Ms. Wankmiller have collaborated on a number of forensic facial image comparison cases, directly leading to Ms. Wankmiller’s current membership in the Facial Identification Scientific Working Group (FISWG) and employment with the Michigan State Police.

Ms. Wankmiller's dissertation research involves bioarchaeological analysis of a pre-Colombian village site in northwestern Costa Rica, combining skeletal analysis with the spatial analysis of mortuary practices. Ms. Wankmiller initially served as Dr. Sauer's assistant on the project in 2007 and has continued to work with the project's archaeologists as well as with archaeologists and staff from the National Museum of Costa Rica. Her successes are a reflection of Dr. Sauer's commitment to forensic anthropology and bioarchaeology as scientific disciplines and his dedicated mentorship in building rapport and maintaining strong collegial relationships with professionals within and outside of Anthropology.

Cate Bird recently graduated from Michigan State University with a PhD in Physical Anthropology under the guidance of Dr. Sauer. Through the analysis of peri-mortem trauma, her research demonstrates that state-sponsored violence in the Soviet Union during the Stalinist period was not performed uniformly across time or personnel. Rather, state violence was implemented at the discretion of individual agents, who differentially complied with state guidelines for execution. Currently, Dr. Bird is developing a regional index of violence which compares peri-mortem trauma across numerous geographical, temporal, and political contexts during the Stalinist period. Dr. Sauer profoundly impacted Dr. Bird's research during the last six years by stressing a holistic, bio-cultural approach to interpersonal and state-sponsored violence. Specifically, he encouraged her to explore not only patterns of trauma at a population level, but also to investigate the social, psychological, and material explanations for this violence in forensic contexts. Dr. Sauer's generalist approach to anthropology has encouraged his students to combine skeletal biology, human rights, and socio-political theory to create robust research agendas.

This presentation demonstrates that strong mentorship can have a significant impact on the quality of the graduate student experience. Dr. Sauer's wisdom, integrity, and personal investment in his students are apparent through generations of graduates. His approach to education serves as a successful example for mentors in forensic anthropology.

Mentor, Forensic Anthropology, Bioarchaeology

H32 An Assessment of Biological Affinity in an Unmarked Cemetery From Nevada: The Importance of Context and Appropriate Reference Samples

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The goal of this presentation is to present the audience with an example of the use of craniometrics to assess biological affinity in an unknown historic population recovered from an unmarked cemetery.

This presentation will impact the forensic science community by highlighting the importance of contextual information, such as spatial distribution and artifactual evidence, and the use of appropriate reference samples when drawing conclusions of biological affinity.

This study presents an analysis of five of the nine individuals recovered in 2000 from an unmarked cemetery in Palisade, Nevada, a ghost town officially abandoned in 1961. The cemetery was discovered during a mining operation, which required removal of the burials. The remains and associated grave items were excavated by local law enforcement and were submitted to the Human Identification Laboratory at California State University, Chico for osteological analysis. These individuals were buried

close by but segregated from the town's main cemetery. This information, coupled with associated grave goods and evidence of poor skeletal and dental health, led to the assertion that these individuals may have been socially marginalized during life. Because the population of Palisade, Nevada, was overwhelmingly White, it was further suggested that this social marginalization could, in part, be due to their ancestral background. Prior craniometric analysis, using the modern reference samples in FORDISC® 3.0, showed definitively that these individuals were of non-European ancestry.¹ The evidence for a particular non-European group or groups, however, was equivocal. This ambiguity may have been a product of inappropriate reference samples. This study tests that assertion by comparing these unknown individuals to known Native American groups from Nevada and other parts of North America.

Because of sample size limitations, males and females were grouped together and craniometric data were scaled using the geometric mean to control for sexual dimorphism. After scaling, craniometric data were aggregated by the group mean of each measurement. Hierarchical cluster based modeling was used to compare the unknown individuals to Great Basin and Western Plains Native American groups (n=43) provided by the Repatriation office, National Museum of Natural History (NMNH), Smithsonian Institution, and Native North Americans from the Howell's sample (n=279).²

The results of the hierarchical cluster analysis show two separate branches, with the unknown individuals being most similar to the Shoshone, followed by the Ute and Santa Cruz groups. The likeness of the unknown individuals with the Shoshone and Ute, who are part of the Northern Uto-Aztecan language family, is not surprising in light of the haplogroup similarities between the unknown individuals (haplogroups B = 50% and C = 50%) and Northern Uto-Aztecan speakers (haplogroups B, C, and D, 42%, 15%, and 43%, respectively; n=116).³ The second branch contains the Paiute, Comanche, Gosiute, Eskimo, and Arikara groups. Given the linguistic and geographic similarities between the Shoshone and Ute and the Paiute and Gosiute, the craniometric difference between these Great Basin groups is surprising.

The results of the current study show that these unknown individuals compare favorably with Great Basin Native Americans, which is consistent with other forms of evidence. These results highlight the importance of contextual evidence and appropriate reference samples when assessing biological affinity. The equivocal FORDISC® 3.0 result from prior analysis appears to be the product of inappropriate reference samples in the Forensic Data Bank for this context. While the Forensic Data Bank contains a Native American sample, this sample consists of modern forensic cases from the American Southwest supplemented with 19th-century Native American remains of undisclosed geographic origin. The 19th-century Native American remains make up over two-thirds of the total Forensic Data Bank sample (44 males, 27 females from a total of 59 males and 32 females).⁴ The fact that these unknown individuals compared more favorably with Great Basin Native Americans than the Forensic Data Bank sample, despite being contemporary with the latter, highlights the range of craniometric variation of North American native groups.

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Ancestry, Craniometrics, FORDISC

H33 Fracture Initiation and Propagation in Pediatric Blunt Cranial Trauma

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After attending this presentation, attendees will gain awareness of: (1) fracture initiation and propagation in pediatric blunt cranial trauma; (2) the applicability of a porcine study as an explanatory model for human forensic cases; and, (3) the implications of this study for the medicolegal interpretation of cranial fracture.

This presentation will impact the forensic science community by addressing misconceptions in current literature regarding the mechanism of cranial fracture that may hinder the forensic investigator's assessment of pediatric cranial injuries.

An accurate interpretation of blunt cranial injury patterns depends on a clear understanding of the mechanism by which cranial fracture is produced. Adult cadaver studies by Gurdjian *et al.* demonstrated that initiation and propagation of cranial fracture depends on impact conditions, namely the shape and kinetic energy of the impacting object.¹ Those studies outlined six distinct impact conditions that influence injury outcomes including type of fracture sustained and the location of fracture initiation either directly at or peripheral to the impact site.

While Gurdjian demonstrated the effect of impact conditions on adult cranial fracture, similar data had not been systematically collected for immature crania. This study utilized an infant porcine model to develop an understanding of the relationship between impact conditions and patterns of pediatric cranial fracture. Single blunt impacts were delivered to fleshed infant pig heads under known conditions. Fracture number, location, and length were assessed for each cranium to characterize the overall injury pattern.

The results of the porcine study revealed peripheral fracture initiation occurring in 100% of 64 impacts involving drops onto a flat, rigid interface. Impacts to the center of the right parietal produced fractures extending from all adjacent sutures both toward and in the opposite direction of the impact site. The effect of this mechanism of fracture initiation was the frequent occurrence of multiple, separately initiating fractures (87.5% of 64 drop experiments), including fractures in bones adjacent to the impacted right parietal (82.8% of 64 drop experiments).

The patterns of injury described in the porcine model have also been observed in several human cases that emerged as part of the Pediatric Cranial Fracture Registry, a National Institute of Justice (NIJ) -funded multidisciplinary effort to establish a national database of pediatric deaths involving blunt force cranial fracture. Of 57 homicides studied, 12 cases exhibited cranial fracture patterns nearly identical to those observed in the porcine model:

multiple, separately initiating fractures with involvement of multiple cranial bones. The striking similarities between these cases and the porcine experiments provides support that: (1) the porcine model has applicability for human forensic cases; (2) peripheral initiation also occurs in human infants; and, (3) the injuries present in these human cases may have been caused by single blows with flat impact surfaces resulting in peripheral initiation.

These results are consistent with Gurdjian's predictions for impacts involving lower energies and flat interfaces. According to Gurdjian, compression of the skull under these conditions produces outward bending in areas peripheral to the point of impact. Because bone is more resistant to compression than tension, fractures initiate at these out-bended areas of tensile stress rather than the impact site. Recent studies by Kroman and others have attempted to discredit peripheral initiation altogether, instead demonstrating initiation at the impact site in adult cadavers.² The current study's results indicate that such a dismissal is premature and misleading, particularly in regard to immature crania.

The understanding of fracture initiation presented by this study has significant implications for the medicolegal interpretation of pediatric cranial injuries. The presence of multiple fractures is generally considered a strong indicator of abuse. If it is assumed that initiation occurs exclusively at the point of impact, it follows that multiple, non-intersecting cranial fractures indicate multiple impacts. However, this study has demonstrated that multiple, non-intersecting cranial fractures may result from a single impact due to fracture initiation occurring peripherally from the impact site. Therefore, the forensic investigator must exercise caution when using such injury patterns to determine abuse in pediatric cases.

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Cranial Fractures, Blunt Force Trauma, Pediatric

H34 Identification of Patterns of Blunt and Gunshot Trauma in a Sample of Alleged Extrajudicial Executions in Colombia

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After attending this presentation, participants will recognize the patterns of trauma and circumstantial factors affecting it when speaking of alleged extrajudicial executions committed in Colombia, South America. Also, participants will have the opportunity to become familiar with some of the issues which forensic professionals have to deal with when operating in a criminal policy-changing scenario, typical of transitional justice processes.

This presentation will impact the forensic science community by exploring a quantitative approach to scientifically understand and evaluate extrajudicial executions in a complex context of ongoing war and sophisticated military strategies.

The goal of this presentation is to introduce the audience to

the conclusions of a research that has been developed by National Center for Human Rights Education in Colombia, regarding the analysis of forensic medical and contextual information of killings alleged to be extrajudicial executions committed between 2002 and 2012. The purpose of this research was to identify the patterns of trauma and contextual variables affecting it as a starting point for future comparisons with *killed in combat* victims.

Since 2008, there has been an increasing interest of local Non-Governmental Organizations (NGOs) to mainstream investigations into extrajudicial executions across the country. A former report documents more than 3,000 killings of civilians by military forces have occurred between 2002 and 2010. This situation revealed the imminent existence of a systematic strategy of some factions of the military to commit the crimes and hide them as legal casualties.

The criminal investigations into those killed have faced several difficulties because of the primacy given to testimonial evidence due to the weakness of forensic science to present significant conclusions based on physical evidence that allow for the distinction between real combat casualties from extrajudicial executions. The lack of understanding on the patterns of trauma and circumstantial factors that characterize both combat killings and extrajudicial executions is the reason such expert opinions about the matter have not been provided.

Thus, this research was carried out to establish a scientific baseline for alleged extrajudicial killings; it was not possible to do that for confirmed extrajudicial executions since there have been only a few cases with a judicial decision and therefore are insufficient for statistical purposes.

A sample of 190 alleged executions was drawn. Collection of data included autopsy, death scene reports, and military reports. Variables for each document were established and information was coded into a relational database management system by using SQL language. Variables regarding autopsy reports included victim profile, cause of death, and mechanism, location, number and lethality of injuries. Variables for death scene report included number of fatal victims, number of survivors, location of the combat, time of the event, and evidence recovered. Variables for military reports included size of combating groups, identification of enemy, and weaponry.

Future research consists of building on a baseline for killings in combat, the comparison of the two samples to identify important similarities and discrepancies, and the construction of a decision tree that can help to establish a route for the evaluation of specific cases.

Combat Trauma, Extrajudicial Executions, Statistical Analysis

H35 Skeletal Characteristics of Quadrilateral Defects in Cranial Bones: A Mixed Mechanism Category of Trauma Associated With Pointed Axes

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After attending this presentation, attendees will have a better understanding of the skeletal characteristics of quadrilateral defects in cranial bones.

This presentation will impact the forensic science community by describing patterned entry and exit wounds in a human rights case and a bioarchaeological sample. These wounds are associated with a mixed mechanism category of skeletal trauma likely caused by a particular class of objects: pointed axes.

This study assesses characteristics of quadrilateral defects on crania from two skeletal samples, including from the Tuskulenai Case and the Battle of Towton. The Tuskulenai case consists of 767 prisoners executed in the Lithuanian Soviet Socialist Republic by the Soviet state from 1944 to 1947. In this study, perimortem trauma was examined in 155 individuals. Twenty-two of these individuals demonstrated 30 quadrilateral entry wounds and 9 quadrilateral exit wounds. Similar quadrilateral defects were observed in remains from the Battle of Towton (A.D. 1461) in North Yorkshire, England. Associated with the Wars of the Roses, this bioarchaeological sample included four individuals who exhibited a total of seven quadrilateral entry wounds. Quadrilateral defects observed in both of these cases were examined with regard to macroscopic characteristics, including entry versus exit wounds, wound shape, wound size, edge damage, and radiating fractures.

Combining the samples, a total of 46 defects were evaluated including 37 entry wounds and 9 exit wounds. Quadrilateral entry defects generally exhibited four linear sides and four 90° angles, resulting in square or rectangular-shaped wounds. However, when accompanied by a high degree of fragmentation, irregular wounds also occurred. The maximum length (mean: 20mm) and maximum width (mean: 16mm) of entry wounds varied considerably, likely due to differing sizes, depths, and angles of penetrating objects. Radiating fractures were observed in 84% of entry wounds and ranged from one to four linear fractures. Finally, all quadrilateral entry wounds demonstrated associated edge damage, including internal beveling or delamination (88%) and external beveling or delamination (87%). Quadrilateral exit wounds were far more variable in size than entry wounds, with only 25% exhibiting square or rectangular shapes. Radiating fractures were observed with all exit wounds and ranged from one to three in number. While internal bevels were limited or lacking on exit wounds, external bevels were present on all exit defects. Finally, crushing, peeling, and lipping were frequently observed on the margins of both entry and exit defects.

This study proposes a class of weaponry, pointed axes, as a primary cause of these quadrilateral defects. Objects associated with these wounds are square or rectangular-shaped in cross-section, have a pointed tip, taper from base to tip, and have sufficient mass to penetrate bone. When impacting bone, these objects represent a combination of sharp force and blunt force trauma. Implements categorized under the class of pointed axes may include pick axes, ice axes, poleaxes, and war hammers.

This study reports the characteristics of quadrilateral entry and exit defects in cranial bones from two skeletal samples. Specifically, quadrilateral entrance wounds were commonly square or rectangular in shape, while exit wounds were more variable in shape and size. While internal and external beveling was observed on entrance wounds, only external beveling was observed on exit wounds. However, both entry and exit wounds exhibited radiating fractures as well additional types of edge damage. Quadrilateral defects likely result from a combination of both sharp and blunt force trauma. Additionally, this study proposes a class of weapon (i.e., pointed axes) commonly associated with these patterned defects. The goal of this presentation is to report a typology of quadrilateral defects and an associated class of weaponry to the forensic science community.

Skeletal Trauma, Quadrilateral Defects, Pointed Axes

H36 The Rorschach Butterfly: The Use of Nomenclature in Lieu of Understanding the Effects and Components of Kinetic Energy in Bone Trauma Interpretations

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After attending this presentation, participants will gain knowledge related to current limitations of bone trauma classifications and terminology used in forensic anthropology with particular reference to blunt and ballistic butterfly fractures.

This presentation impacts the forensic science community by contributing to knowledge of bone trauma classifications and use of misleading or misdirected nomenclature associated with fracture pattern recognition. Blunt, sharp, and ballistic (contra "projectile" in much of anthropology) trauma categories are used to distinguish and interpret fracture characteristics in clinical settings and for description in a court of law. Bone trauma categories must convey bones' response to overall absorption of kinetic energy ($KE = mv^2/2$) in which velocity may be a recognizable feature. However, anthropologists continually use the term "butterfly" to explain fracture shape and propagation as well as extrapolate external loading conditions in both blunt and ballistic long bones injuries.

In 1885, Messerer noticed that bending stresses on a tibia caused distinct wedged-shaped fracture patterns that at the time was also considered useful in defining direction and location of the externally applied force. More recently, a *Messerer fracture* primarily appears in forensic pathology literature and is applied to very specific injury-producing events, particularly pedestrian-vehicle accidents.¹ Yet his pioneering work on fracture pattern recognition in bending bone is ubiquitous in biomedical and forensic literature and is commonly referred to as butterfly fracture/fragments, tension wedge-fracture, and bending wedge-fractures. With an extensive research history of fracture propagation and bending bone, the association of "a bending butterfly pattern" in ballistic injuries is incorrect and is apparently associated with a dual-meaning for a butterfly fracture in the academic literature. For example, in 1915, La Garde described a perpendicular gunshot wound to a tibia (plug and spall and associated radiating fractures) as a butterfly fracture; his observations brought forth the use of current terminology such as ballistic, false, and/or double-butterflies.²

Biomechanically, blunt and ballistic butterfly fractures are not interchangeable in recognition or interpretation. Since the biomechanical response and fracture mechanics of bone are dependent on the external loading conditions (load-rate, contact area, impact location), butterfly descriptors are different for each situation and are essentially opposite patterns. The important question to answer with these various butterfly configurations is whether the morphological descriptors are useful in understanding fracture propagation and subsequent interpretations of fracture etiology as well as direction of failure.

The purpose of this presentation is to redirect anthropologists from relying on terminology to describe energy and focus on interpreting, at least at the basic level, the biomechanics behind bone fracture. To achieve this goal, lesion characteristics of blunt and ballistic butterfly fractures are described and contextualized, related to soft tissues and within a basic biomechanical framework. Case studies are used to illustrate the need for forensic specialists to examine external loading conditions through closer examinations of all fractured surfaces (macro and microscopically) as well as the entire pattern of trauma on a body, including the associated damage on skin and soft tissues whenever possible, prior to presenting specific fracture interpretations.³

Academics are urged to go beyond comfortable pedagogical practice and to explain modes of failure, tension, compression, and shear, along with varied nomenclature as a means to improve understanding and facilitate the maturation of bone trauma as a scientific discipline. A butterfly fracture is often an anthropology student's first exposure to the biomechanics of bone trauma but merely recognizing a butterfly fractures does not lead to success in bone trauma interpretations, as the biomechanical response of bone to an external loading condition is far more important than the various names used to describe a broken bone or for a bone to be thrown into a generic category.

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Classification of Bone Trauma, Biomechanics, Butterfly Fractures

H37 Postmortem Smoothing of Fractured Fresh Bones in Bodies Immersed in Moving Water: Forensic Cases Aid in Trauma Interpretation of an Archaeological Skeleton

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After attending this presentation, attendees will understand the effect of moving water on bodies, with an emphasis on broken bones subjected to bone-on-bone abrasion. Interdisciplinary collaboration between forensic and archaeological osteologists is highlighted as is the potential for forensic anthropology to contribute to archaeological taphonomy and trauma interpretations.

This presentation will impact the forensic science community by raising awareness of the fact that forensic anthropology is fundamentally an applied field, drawing inferences from events surrounding death, deposition, and recovery using methods developed in archaeology, physical anthropology, forensic pathology, and geology. In contrast, the contributions of forensic research to the accurate interpretation of archaeological death assemblages are less often acknowledged. This is unfortunate because forensic research has the unique advantage of confession or witness testimony to confirm inferences drawn from the skeleton about the circumstances of death. Documented forensic-related findings can be applied to archaeological or paleontological studies of the remote past where all that remains is the death assemblage. Here, taphonomic patterns in two forensic cases are consistent with the postmortem modification of a bone from a prehistoric skeleton. Forensic-derived information contributes to a more informed interpretation of the context of the death that took place hundreds of years ago.

The archaeological skeleton is from Norris Farms #36, a cemetery associated with a small village in Illinois dating to ca. AD

1300. The woman of interest was one of several people killed in a series of attacks.¹ The left ulna of this well-preserved skeleton was fractured and the originally sharply defined, broken edges were worn smooth. As the individual's skeleton was archaeologically recovered in anatomical order, the polishing must have occurred when soft tissue held the bone fragments in place. How the adjoining edges became polished remained a mystery until similar forms of abrasion were seen in recent forensic cases.

Two Mercyhurst University water-recovery forensic cases had abrasion patterns similar to what was observed on the Norris Farms skeleton. The bones of a leg displayed peri-mortem fractures polished by bone-on-stones abrasions and even one case of bone-on-bone smoothing, where there was evidence of a sharp, fractured fibula edge moving against the adjacent tibia. The bones were all held in place by soft tissue and a boot. Moving water turbulence, such as the Niagara and Allegheny rivers where these individuals were found, was sufficient for enough bone-on-stones and bone-on-bone movement to smooth and etch the adjacent shaft. Moving water was likewise probably responsible for the smoothing seen on the adjoining ends of the broken ulna in the archaeological skeleton.

The effects of fluvial action in terms of damage to exposed bone and on assemblage composition are well described in the literature.² While conducting this study, however, this particular form of bone-on-bone abrasion, which occurs when soft tissue is still present, was not found to have been reported. Although the circumstances of death were different for the forensic cases and the archaeological skeleton, the taphonomic patterns indicate that what happened afterwards — bodies were immersed for prolonged periods in moving water — was the same. In the absence of documented forensic cases, it was not possible to identify how the adjoining broken ends of the ulna in the archaeological skeleton were smoothed. Knowing the circumstances under which such bone-on-bone or bone-to-abrasive materials occurs, allows us to identify where that individual died and to infer that women spent time in the wet Illinois River floodplain where they were vulnerable to attacks. Other such skeletons probably exist in archaeological collections, but the significance of the smooth edges of fractured bones is unrecognized.

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Bioarchaeology, Skeletal Trauma, Norris Farms

H38 Distinguishing Homicides and Suicides in Firearm Fatalities: The Role of Skeletal Trauma Analysis

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After attending this presentation, attendees will understand how skeletal gunshot trauma analysis can provide information for identifying manner of death through consideration of four specific factors: (1) anatomic location of entrance wound; (2) bullet direction; (3) number of shots; and, (4) fracture severity/patterns.

This presentation will impact the forensic science community by identifying new ways in which firearm fatalities and manner of death can be investigated through the use of skeletal trauma analysis. Specifically, it presents new findings that broaden the scope of anthropological research on skeletal gunshot trauma and uniquely represent the important role of the anthropologist in the forensic science community.

Although it is not the anthropologist's responsibility to determine manner of death in a forensic case, an expertise in osteology and the biomechanics of high velocity projectile trauma to bone may contribute to the pathologist's final conclusions about manner of death in firearm fatalities. Previous research looking at variation between homicides and suicides in firearm fatalities has primarily used soft tissue trauma analysis and autopsy reports to identify common characteristics specific to these manners of death.¹⁻⁹ They have found that anatomic location of entrance wound, bullet direction, number of shots, and range of fire are important factors in understanding this variation. Anthropological research has not explored how this variation is expressed on the skeleton and knowledge of these differences can better equip anthropologists to report pertinent information that can lead to accurate determinations of manner of death. The current study explores the factors of anatomic location of entrance wound, bullet direction, and number of shots, with additional consideration to fracture severity and fracture patterns, in place of range of fire.

The William M. Bass Donated Skeletal Collection and the Antioquia Modern Skeletal Reference Collection provided a total sample of 15 suicide and 19 homicide cases, totaling 16 suicidal and 35 homicidal gunshot wounds. Each factor observed was analyzed to identify statistically significant differences between homicides and suicides.

This research found that localized entry sites to the front and right sides of the head and to the chest were common in suicides. Entries to the left and front sides of the head and to the posterior side of the head and body were common in homicides. Further analysis of sequence of shots and entry sites revealed that the front of the head, common in both homicides and suicides, was more common in secondary gunshot wounds in homicides. Bullet directions common in suicides were right to left through the sagittal plane, and anterior to posterior through the coronal plane. The left to right direction through the sagittal plane was most common for homicides, whereas the anterior to posterior and posterior to anterior directions through the coronal plane were almost equally represented in homicides. Further analysis of sequence of shots and bullet direction revealed that the anterior to posterior direction was more common in secondary gunshot wounds in homicides. The transverse plane did not show statistically significant differences between homicides and suicides for either the inferior to superior or superior to inferior directions. For number of shots, homicides more commonly expressed multiple gunshot wounds and suicides more commonly expressed single gunshot wounds. Fracture severity analyses revealed that the presence of tertiary fractures (concentric fractures) in entrance wounds was more common in suicides. The presence of secondary fractures (radiating fractures) as the most severe fracture in entrance wounds was more common in homicides. These results suggest that fracture severity is higher in suicides, based on the amount of kinetic energy dispersed at impact, although it is unclear what factor influences these differences (range of fire, type of firearm, caliber of bullet, etc.). There was also a significant difference between homicides and suicides in fracture patterns for both entrance and exit wounds.

These findings are based on a small sample and should be considered with caution, especially for use in a forensic setting. Further research is crucial to better understanding the results of this study. Specifically, sequencing of shots should be further explored for its utility in distinguishing homicides and suicides as well as providing a better understanding of multi-shot cases. Additionally, research should be conducted on fracture severity and fracture patterns with specific consideration to factors that influence the variation seen between homicides and suicides.

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Skeletal Gunshot Trauma, Manner of Death, Fracture Severity

H39A Rare Case of an Intact Bone Plug Associated With a Gunshot Exit Wound

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The goal of this presentation is to report on the findings of a rare case of an intact bone plug associated with ballistic trauma to the cranium of an adult male exhumed from a human rights case in Lithuania.

This presentation will impact the forensic science community by addressing variations in manifestations of gunshot wounds in skeletal remains and the continued importance of precise archaeological excavations of mass graves.

Between September 1944 and April 1947, 767 individuals were executed by the Soviet security police in Vilnius, Lithuania. After the executions, victims were buried in clandestine mass graves at the former Tuskulenai Estate, a former palace from the 16th-century located on the outskirts of Vilnius. The Republic of Lithuania regained its independence from the Soviet Union in 1990, and in 1993 Soviet security documents were discovered

indicating the presence of mass graves on the Tuskulenai Estate. Excavations of the estate in 1994, 1995, and 2003 exposed 45 mass graves, each containing between 1 and 154 individuals. Skeletal analysis revealed a total of 724 individuals, including 720 males and 4 females. Approximately 97% of these victims demonstrated evidence of peri-mortem trauma, which included gunshot wounds, sharp force trauma, blunt force trauma, or a combination of these traumatic mechanisms.

In 2012, skeletal trauma in 155 individuals in the Tuskulenai case was analyzed. During this analysis, individual number 626, an adult male, was found to have a single, execution-style gunshot wound to the left occipital with a posterior-anterior trajectory. Both an entry and exit wounds were observed, and a deformed bullet was recovered with the remains. The circular gunshot entrance wound measured 6 millimeters in diameter and demonstrated typical internal beveling associated with the entry of a ballistic projectile. An associated exit wound was observed on the right frontal bone. This circular exit wound measured 9 millimeters in diameter and displayed a broad, external bevel.

Additionally, a disarticulated, but intact plug of bone belonging to the exit wound was recovered with the remains. The circular bone plug measured 20 millimeters on its ectocranial surface and 9 millimeters on the endocranial surface. When re-articulated, this plug fit securely into the exit wound. Four small linear fractures extended through the external surface of the plug, but these fractures did not extend to the internal surface. This fracture is consistent with the bone failing on the tensile side first but not on the compressive side. The projectile likely lost its ballistic energy, causing plastic deformation at the exit wound site, where the initial failure occurred on the external surface. This fracture pattern is more consistent with blunt force trauma than with ballistic trauma.

This study presents an unusual case of gunshot trauma observed on the skeletal remains of an executed individual from a post-World War II human rights context. The bullet entered the cranial vault posteriorly, lost its ballistic energy, and caused plastic deformation at the exit site, consistent with blunt force trauma. In turn, the displaced bone plug associated with the exit wound failed on the tensile side of force, but not on the compressive side, permitting its complete recovery. This study contributes to the forensic community by providing an in-depth examination of alternative manifestations of typical plug and spall formations associated with gunshot exit wounds in bone. Additionally, this study demonstrates the necessity of systematic archaeological excavations of mass graves. Without meticulous excavation, both the bone plug and the bullet might not have been recovered from the grave.

Gunshot Wound Trauma, Plug, Mass Graves

H40 A Case Study of Glacial Taphonomy

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After attending this presentation, attendees will understand the unique taphonomic signature that can present on human remains recovered from a glacial environment.

This presentation will impact the forensic science community by defining taphonomic processes that are encountered in a glacial setting.

In 2012 and 2013, the Joint Prisoners of War, Missing in Action Accounting Command Central Identification Laboratory (JPAC-CIL) lead recovery efforts of a 1952 aircraft crash of a C-124

carrying 52 individuals on Colony Glacier in Matanuska-Susitna Borough, Alaska. The initial crash site was recorded approximately 18km east of where the JPAC-CIL recovered the majority of human remains, aircraft wreckage, and other associated artifacts. This case presents a unique opportunity to investigate the taphonomic effects related to the deposition and subsequent movement within and across the ice of human remains that have been subject to glacial processes for a known period of time. The taphonomic processes affecting the remains can be summarized as follows: the initial aircraft crash; subsequent snow burial of wreckage and remains; movement of remains and other associated artifacts through and/or on the ice; melting of the ice and snow leading to the exposure of the remains and artifacts; and the recovery of these items by the JPAC-CIL. Each of these events contributes to the unique taphonomic pattern apparent on these remains. The primary taphonomic features described on human remains recovered from this incident are associated with the movement within and across the ice and include abrasion, shearing, fraying, and splitting. These unique taphonomic characteristics are unlike other taphonomic effects that can result from similar environments (such as cold climates).

Global climate change forecasts indicate glaciers will continue to recede in the coming decades. It is likely that more human remains cases may be exposed from previously inaccessible areas as ice and snow cover disappear. The unique taphonomy related to glacial deposition and movement may aid investigators in recognizing the contextual history of human remains recovered in these types of environments.

Taphonomy, Glacier, Aircraft Crash Recovery

H41 Comparing Feral Pig and Coyote Scavenging and Dispersal Patterns in the Greater Yosemite Ecosystem in California

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After attending this presentation, attendees will have increased knowledge of feral pig (*Sus scrofa*) and coyote (*Canis latrans*) scavenging and dispersal patterns when the two species feed concurrently.

This presentation will impact the forensic science community by documenting feral pig and coyote scavenging in a natural setting, which can be used to suggest best practices for law enforcement officers searching for human remains in areas where feral pigs and/or coyotes are common.

Individuals left to decompose in outdoor environments may be subjected to all manner of carnivore scavenging before they are recovered. This study documents scavenging patterns to determine the effect of multiple species on human-sized pig (*S. scrofa*) remains in an outdoor setting. It is hypothesized that the interactions of multiple species of scavengers modifying the same remains will impact the dispersal of skeletal elements differently than in studies documenting single scavenger taxa.

Research was conducted on private land in the Yosemite Valley, California, USA. Six study pigs of comparable mass to adult humans were left exposed for five weeks beginning May 22, 2012, and observed daily for the first two weeks. After five weeks, one of the study pigs had been removed and the remaining five had become mummified, although some disarticulation was observed. The bones that had been moved were mapped, collected, and examined for scavenger modification. Approximately one year

after the completion of the study, the original site of deposition was revisited, and any bones from the five remaining mummified study pigs were mapped and collected.

Results of this study suggest that regular human interaction has an impact on carnivore behavior. During the first two weeks of the study when the remains were being directly observed, carnivore activity was non-existent. Two days after daily observations had ceased, coyotes and feral pigs began scavenging the remains. Coyotes tended to scavenge those study pigs closest to their game trails and the dry creek bed they used for travel, while feral pigs showed no preference for location, often moving from one set of remains to the next during the course of a feeding session.

Coyotes were responsible for removing the study pig remains from the site during the initial five weeks of the trial and disturbed an additional two sets of remains during the course of scavenging. Bones located outside of the immediate area of original deposition exhibited punctures and pits indicative of canid gnawing. Remains that were scavenged exclusively by feral pigs during the trial period were largely undisturbed apart from expansion of the openings in the neck and abdomen originally created by decomposition. Parallel scoring as a result of the incisors was also found on long bones associated with remains scavenged by feral pigs, in accordance with other researchers.

When the site was revisited approximately one year after the research began, small bones such as vertebral centra, ribs, and epiphyses as well as a few long bones and a mandible could still be found in the original placement area. These bones were the only visible evidence that remains were deposited in the area.

Analysis of dispersal patterns revealed that a majority of skeletal elements were found either adjacent to the original deposition site or along nearby game trails. This suggests following game trails is an effective technique for law enforcement attempting to locate remains. However, entire carcasses can be removed with minimal skeletal elements or soft tissue left behind, as was observed with the first study pig removed from the site. Coyotes were the main agents of bone destruction and dispersal, although feral pigs did modify the remains in telltale ways. The results of this research stress the importance of detailed searches for any remaining bones, which can be used to establish the original site of deposition even a year later in outdoor crime scenes with known or suspected scavenging of remains.

Taphonomy, Scavenging, Yosemite

H42 The Postmortem Fate of Anthropometric Measurements: Taphonomic Alteration of Landmarks in Buried Skeletal Remains

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After attending this presentation, attendees will understand how skeletal remains recovered from burials present taphonomic alterations which affect the availability of anthropometric measures differently than anatomical collection's samples.

This presentation will impact the forensic science community by suggesting what measurements are more frequently preserved in buried skeletal remains and so are more useful to create applicable discriminate functions for anthropometric evaluations.

The analysis has been conducted on a sample of 644 historical buried skeletons of adults (376 males; 268 females) from Italy. All the individuals were buried supine in earthy pits or large

stone coffins; the context did not show any peculiar environmental condition that could influence the bone preservation or incremental lack of any specific anatomical region.

Due to its complexity, the skull was divided in seven different regions: basis; neurocranium; calva; splanchnocranium; orbital region; nasal region; palate; and, mandible. This division is based on anatomy of performing measurements for each specific area. Long bones and scapula are studied without specific internal divisions. A selection of 120 measurements, classified by a new coding system, has been sorted according Martin-Saller standards.^{1,2}

A preliminary analysis demonstrated that a complete skull, in which all measurements (57) can be collected, represents less than 1% of the whole sample, while every single area appears more frequently accessible for anthropometric purposes: basis 25%; calva 23%; orbital region 17%; nasal region 18%; palate 19%; and, mandible 14%.³ The combination of several areas leads to worse results: neurocranium 8% and splanchnocranium 7%. These data do not seem to be predictable and do not subordinate to any specific taphonomic law.

The analysis of the preservation of every single cranial measurement demonstrates that a third of them can be read in more than 50% of the whole sample, while only nine are readable in less than 25% of the cases. The more preserved measurements are the mandibular ones, where six out of ten are readable in more than 50% of the samples and only one is readable in 31%. The worst preserved landmarks are in the nasal area, where two measurements are readable in 33% of the cases and the other only in 22% (nasal-malar cord) and 16% (nasal-malar breath).

A new more extensive study on post-cranial long bones detected a uniform trend where the metaphyseal landmarks are the most frequently traced (usually 80%), followed by epiphyseal ones (average 50%). More affected by diagenetic phenomena are the lengths that are often bordered by landmarks placed on fragile epiphysis and processes. A relevant trend detected shows a differential measurement preservation for gender and laterality: male and right side samples indeed seem more undamaged, presumably in relation to their increased resistance resulting from sexual and functional characteristics.

Particularly dramatic is the condition of the scapula's body conservation (measures a readable average in less than 5% of the cases), probably due to its remarkably thin and fragile structure. Also the anatomical position of this bone can increase its taphonomic destruction: in a supine body, it could be more subjected to diagenetic phenomena due to stagnation in moisture and decomposition fluids. The better results observed in the glenoid cavity (preserved on average in 75% of the cases), morphologically and topographically without the issues illustrated for the scapular body, confirm this assumption.

This presentation will illustrate how the cranial measures can seldom be used for statistical analyses due to poor taphonomic resistance of their landmarks; considering the predominance of some single measurements or small cranial zones, a specific strategy for each anatomical unit or individual measurement is suggested. According to their frequency, the postcranial measurements (particularly midshaft diameters and circumferences) are more appropriate to create usable discriminant functions for biological profile analysis.

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Anthropometry, Statistical Analysis, Taphonomy

H43 Microscopic Residues of Bone From Dissolving Human Remains in Acids

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After attending this presentation, attendees will understand that it is possible to dispose of human remains by dissolving them in acids, but that it is nearly impossible to do so without leaving microscopic residues.

This presentation will impact the forensic science community by showing which kind of microscopic residues will remain after dissolving human remains in acid and how these residues can be detected.

Dissolving bodies in acids is a well-known method of disposing of human remains and has been practiced throughout the years. History's most notorious case is the "acid bath murderer" John Haigh, an English serial killer during the 1940s, who was convicted and subsequently executed for murdering six people. He dissolved the bodies in concentrated sulfuric acid, believing that if the victims' bodies could not be found, then a murder conviction would not be possible.

During the last decade in the Netherlands, two cases have emerged in which human remains were treated with acid. In the first case, the remains of a burned body were treated with hydrofluoric acid. In the second case, two complete bodies were dissolved in a mixture of hydrochloric and sulfuric acid.

In the first case, a witness declared that a suspect involved in drug trafficking had killed his missing companion and burned him in an improvised incinerator. However, no remains of either the victim or the incinerator were found. Later, during another house search, an off-white concrete-like object, speckled with pink and brown spots, was found buried in a refuse bag in the suspect's garden. At first, no one had an idea what this odd-appearing material could be. After preparing a polished section of the material in question, its internal structure was revealed. The white material consisted of gypsum, and the pink and brown spots consisted of some sand and thin-walled structures containing calcium, phosphorus, and fluorine. Because of this odd composition, it was thought that these mysterious thin-walled structures were originally bone that had been almost completely dissolved by hydrofluoric acid alone or a mixture of hydrofluoric and other acids.

In order to confirm this hypothesis, a series of experiments were launched, in which cremated bone was exposed to mixtures of acids of different strengths and compositions. As the suspect was a professional welder, the experiments focused on pickling acid and pickling paste, which contain a mixture of hydrofluoric and nitric acid. The experiments revealed that it was possible to reproduce the thin-walled structures; the more diluted the acid was, the more delicate the thin-walled structures became.

In the second case, a conscientious witness declared to the police that he helped to dispose of two bodies by dissolving them in a mixture of hydrochloric and sulfuric acids in a plastic barrel. The mixture was stirred regularly and floating fatty residue was skimmed off and drained in a sink. A portion of the broth was regularly replaced by fresh acid. Solid substances from this

removed broth were set apart and treated separately in a bucket of fresh acid. After two weeks, when the bodies were dissolved, the remaining fluid was poured down the drain adjacent to the witness' house.

The contents of the drain and the sewers were secured by the police and consisted of several kilograms of waste, including sand, demolition materials, and stone. After carefully sifting through the contents of the drain, one pivot tooth, a small piece of epidermis, and four residual particles with the same elemental composition as bone (calcium and phosphorus) were found. The four remnants were subjected to DNA-analysis but unfortunately it was not possible to obtain a DNA profile of one of the victims.

Apart from the macroscopic findings, in almost every sample, microscopic residues of bone were found. These microscopic residues fall into three categories: (1) bone, partly acid-digested at the edges; (2) thin-walled structures such as those described above; and, (3) recrystallized calcium phosphate. Although some may believe it is possible to dissolve a body in acid completely, at least some of the aforementioned microscopic residues will always be found.

Bone, Acid, SEM/EDX

H44 Impact of Embalming and Burial on Decomposition

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The goal of this presentation is to assess how the common process of embalming affects decomposition rates in both buried and surface body deposits and how embalming affects Volatile Fatty Acid (VFA) concentrations and leaching into surrounding soils. It also reveals the degree to which necrophagous insects are attracted to embalmed remains in the humid sub-tropical climate.

This presentation will impact the forensic science community by expanding the knowledge available concerning postmortem interval in Kentucky, as well as the postmortem fate of the embalmed, and will lend itself in disaster situations and human rights issues by distinguishing recent deaths from those remains that have received formal mortuary preparation. The study also sets a baseline for determining how far from the body volatile fatty acids are leached from both embalmed and non-embalmed bodies, thereby allowing clandestine graves or disinterments to be discovered through soil analysis.

Since VFAs can be detected in soil, they have the potential to yield a wealth of information concerning clandestine graves, secondary burial sites, archaeological data, and overall soil composition. For example, archaeological sites often contain volatile compounds from plant matter that was deposited during settlement. Excavations at such sites frequently reveal the presence of volatile compounds long after an organism has died and the site has been abandoned. Also, in forensic settings, human volatile fatty acids have been detected in a cemetery in Duz, Kosovo, as well as a mass grave site in Knin, Croatia, as many as six years after the bodies had been buried.¹

The present study took place in Kentucky and used six still-born fetal pigs (*Sus scrofa*) to examine the decomposition differences and the leaching of volatile fatty acids between both non-embalmed and embalmed remains deposited on the surface, as well as remains buried at depths of two feet and four feet. This study reports the concentrations of volatile fatty acids leached away from the body in the surrounding soil on both a horizontal plane and a vertical plane.

Results of the analysis are reported in parts-per-

billion for valeric, iso-valeric, butyric, and iso-butyric acids. The soil immediately below the surface pigs did not retain as high concentrations of VFAs as did their buried counterparts. In general, buried remains tend to retain higher VFA concentrations than surface remains due to the water solubility of VFAs. For example, the soil directly below the embalmed pig buried at two feet had 181, 254, 481, and 727 parts-per-billion (ppb), while the soil immediately below the embalmed surface pig had 0, 9.6, 46, and 42 ppb, respectively. The soil associated with the embalmed and non-embalmed surface pigs contained three of the four acids but had very low concentrations of butyric acid. Buried specimens retained higher levels of all four acids tested. Additionally, soil associated with the embalmed remains yielded the highest concentrations of all four acids, especially in the soil directly surrounding the pig.

This study found that indeed volatile fatty acids are leached on both horizontal and vertical planes, though the concentrations tend to be higher in a vertical direction but also tend to follow the slope of the land. Finally, the practice of embalming does not seem to degrade the concentration of volatile fatty acids. Surprisingly, embalming actually seems to increase or preserve VFA concentrations.

Finally, this study also demonstrates that insects are not equally attracted to embalmed versus non-embalmed remains. A delay of approximately 24 hours occurred in blowfly arrival at the embalmed remains, while the unembalmed remains attracted blowflies within minutes of deposition. Overall, the unembalmed remains hosted more insects (especially blowflies) over a short period of time, while the embalmed remains attracted fewer blowflies and more beetles for a period of over 30 days. Buried remains lacked any evidence of insect activity at the time of exhumation 153 days later.

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Forensic Anthropology, Embalming, Volatile Fatty Acids

H45 The Effect of Clothing on the Rate of Decomposition and Insect Colonization on *Sus Scrofa* Carcasses

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After attending this presentation, attendees will be able to understand how the presence of clothing affects the rate and pattern of decomposition on pig carcasses. Furthermore, attendees will be aware of any differences in the appearance and colonization preferences of forensically relevant insects between clothed and unclothed remains.

This presentation will impact the forensic science community by increasing awareness as to what extrinsic factors significantly alter the rate of decomposition and how these factors may influence the overall pattern of decomposition.

Although the presence of clothing on decomposing human remains is frequently encountered in forensic cases, there have been few quantitative studies regarding clothing's effect on

decomposition.¹ Several retrospective studies and case reports have claimed that clothing retards the rate of decomposition because it both prevents insects from accessing desirable locations for oviposition and helps to protect decomposing tissue from external factors (e.g., sunlight or scavenging).¹⁻³ Conversely, other experimental studies noted accelerations in the rate of decomposition proposing clothing creates new areas for insect oviposition and reduces tissue desiccation.⁴ Lastly, research has suggested that clothing has no quantitative effect on the rate of decomposition.⁵ The inconsistent nature of these findings was the impetus for a new, quantitative study to examine the effect of clothing on rate of decomposition and insect colonization preferences.

Ethical approval was obtained for the use of 20 domestic pig (*Sus scrofa*) carcasses, which were dispatched and transported to the Taphonomic Research in Anthropology Center for Experimental Study (TRACES), a research facility located in Northwest England owned by the University of Central Lancashire (UCLan). The carcasses were separated into two groups clothed or unclothed and carcasses were dressed in identical white cotton t-shirts, black cotton boxer shorts, and belts in order to mimic loose-fitting summer clothing.

Data collection occurred at regular accumulated degree day intervals; the level of decomposition, pattern of decomposition and Diptera present were documented. Results indicated a statistically significant difference ($t_{427}=2.59$, $p=0.010$) between the decomposition rate of unclothed and clothed carcasses. This result must be interpreted with caution, however, as it equates to marginal scoring changes that become irrelevant in terms of forensic practicality. The overall decomposition rates from each carcass group are too similar to separate when applying a 95% Confidence Interval, which means that, although statistically significant, from a practical forensic point of view they are not sufficiently dissimilar as to warrant the application of different formulas. In regard to the overall pattern of decomposition, the carcasses appeared to follow a different pattern than what has been reported previously and differences appeared qualitatively between the two carcass groups which could indicate colonization preferences for insects.

In summary, the results of this study revealed that the presence of loose-fitting summer clothing does not affect the rate of carcass decomposition within a practical forensic context. Clothing did, however, provide colonizing insects with new areas for oviposition which resulted in differing localized patterns of decomposition between unclothed and clothed carcasses.

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Clothing, Decomposition, Pattern

H46 Regional Taphonomy of the Southeast United States: The Role of Algae in the Degradation of Diagnostic Traits of Sharp Force Trauma

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After attending this presentation, participants will better understand the effects of fresh water microflora on the degradation of diagnostic traits of Sharp Force Trauma (SFT) on bone.

This presentation will impact the forensic science community by demonstrating how the microflora specific to a southeastern fresh water environment can contribute to the taphonomic condition of bone.

The microenvironment in the depositional context of skeletal remains is important in reconstructing peri- and postmortem events and determining the postmortem interval. Taphonomic condition can be influenced by a wide variety of confounding factors, which have been demonstrated to vary by region. The scavenger guild impacting the rate and pattern of decomposition will differ by region, including not only mammalian and avian scavengers, but also insects and microscopic organisms.¹ Remains recovered from aquatic environments are exposed to a unique set of taphonomic agents. Oceans, rivers, lakes, and ponds may differ radically with respect to temperature, depth, salinity, oxygenation, current, and life forms.² Each of these factors affect the taphonomic condition of skeletal remains in significant ways, and may impact the appearance of diagnostic characteristics of traumatic injury.

This research examines the effects of algae, which is one contributing factor prevalent in Southeast taphonomy. It has previously been proposed that bacteria functions as a degradative agent, causing bone to decompose at a faster rate than bone which is not exposed to bacteria.³ Although algae do not consume bone, there is some anecdotal evidence that algal growth also speeds the breakdown of skeletal elements. This study hypothesizes that it is not the growth of simple autotrophic organisms on skeletal remains that results in increased rate of bone degradation in fresh water contexts, but rather that the destruction is due to the action of aquatic organisms feeding on the algae. Foot bones of *Sus scrofa* (domestic pig) were cut with an identical tool multiple times at various depths to simulate defensive wounds and each cut mark was photographed and measured. Cortical condition was examined and photographed under a microscope to record density prior to exposure to the experimental conditions. The test specimens were then submerged in water at the edge of a lake in rural western Georgia, while others were submerged in water collected from the same lake but maintained in the lab to protect the specimens from the actions of other organisms. Control specimens were submerged in distilled water in the lab. Over a period of five months, the samples were examined and photographed on a weekly basis. Water in the lab conditions was drained and replaced weekly, and samples of the lake and lab water were examined by a biologist to determine the presence and number of microorganisms. The cortical surfaces and cutmarks were then reexamined and photographed, measuring the visibility of the cuts. The effects of the microflora on the final condition of the cortex and cut marks was then evaluated.

Isolating the effects of specific taphonomic agents is critical to understanding regional taphonomy. This study examines the effects of exposure to algae in aquatic environments in the Southeast and attempts to explain why autotrophic organisms can affect the condition of bone.

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Sharp Force Trauma, Taphonomy, Algae

H47 Evaluating the Use of Accumulated Degree Days and Total Body Score to Estimate Time-Since-Death of Human Remains in Central Texas

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After attending this presentation, attendees will better understand how the extreme heat, radiation, and arid environment of Central Texas affects the use of current total body score methods to estimate time-since-death.

This presentation will impact the forensic science community by elucidating the need for a more regional approach to time-since-death estimation based on an evaluation of current anthropology methods using human remains in Central Texas.

The ability of a forensic anthropologist to offer law enforcement an accurate time since death or Postmortem Interval (PMI) estimate is integral to a successful relationship between law enforcement and the field of anthropology. Often, a PMI estimate is the only aspect of a forensic case in which anthropologists are asked to participate. The ability of anthropologists to estimate PMI, therefore, must be improved and current methods must be evaluated in order to meet the new standards of practice as outlined by Scientific Working Group for Forensic Anthropology (SWGANTH).

This study evaluated the Megyesi *et al.* method for estimating PMI from human remains found in the summer in Central Texas.¹ To date, the Megyesi method is the only widely accepted method for quantitatively estimating PMI of human remains. The method was developed based on photographic observations of forensic cases from across the United States, but mostly concentrated in both Indiana and Illinois. The climate in Central Texas, however, varies greatly from the climates of both Indiana and Illinois. Central Texas is extremely dry and humid with high levels of solar radiation. Average temperatures for the year are in the low 80s Fahrenheit, with summer temperatures plateauing around 100° Fahrenheit for weeks at a time. The objective of this study, therefore, was twofold. First, the aim was to test the method's accuracy when estimating PMI from human remains in Central Texas. Second, if the method proved inaccurate, the aim was to evaluate whether the variables outlined in the method accurately described the decomposition process in Central Texas.

The study evaluated the decomposition of ($N=43$) donations made to the Forensic Anthropology Center at Texas State (FACTS) in San Marcos, TX. All of the donations were human subjects placed at the facility between 2012 and 2013 with a known PMI of less than one year. All of the donations were evaluated in the month of July 2013, and all PMI estimates were calculated from

the date of placement at the facility. In some cases, the date of death preceded the date of placement by as much as two weeks. Photographs were taken to document the scoring and the amount of shade versus sun was recorded. Following the original method, Total Body Score (TBS) was calculated for each donation. From the TBS, the Accumulated Degree Days (ADD) and 95% confidence interval range were calculated for each donation following the Megyesi method. Accuracy was calculated by comparing whether the date of placement fell within the 95% confidence interval range for each donation's ADD. Qualitative assessments of the Megyesi variables were described by comparing the TBS of each donation to the photographs taken during scoring and the expected description corresponding to the TBS scores listed in the original method.

Overall, the method performed poorly for estimating PMI from human remains in Central Texas. Only five donations fell within the 95% confidence interval of the Megyesi method. The qualitative assessment of Megyesi's variables revealed that, overall, her descriptions of the TBS stages matched the TBS calculated for this study. The qualitative assessment of the TBS scores for this study, however, revealed that in Central Texas there could be a separate stage for mummification compared to Megyesi's incorporation of mummification into the advanced stage. Future research using modifications to Megyesi's method will be discussed as well as the need to test current PMI estimation methods across the United States in varying environments.

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PMI, Time-Since-Death, Human Decomposition

H48 Characteristics of Penetrating Screwdriver Trauma to the Cranium

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After attending this presentation, attendees will gain an understanding of screwdriver trauma to the crania and the potential for injury characteristics to be used to ascertain weapon type.

This presentation will impact the forensic science community by presenting results from an initial study of screwdriver trauma to the cranium and reporting on wound characteristics typical of a specific screwdriver type. The findings of this study may allow screwdriver type to be inferred from wound characteristics.

Data from the Federal Bureau of Investigation (FBI) indicates that 13% of 68,720 homicides committed between 2007 and 2011 involved knives or cutting instruments.¹ Matching an injury to an implement by tool mark analysis is an important element of any forensic investigation involving sharp force trauma. Previous works have mainly concentrated on knife and saw marks.^{2,3} Even though homicide cases with screwdrivers are rare, screwdrivers have been reported as weapons used for fatal stabbings.⁴ A screwdriver is easily concealable due to its small size and weight and easily obtainable.

Twenty pigs' (*Sus scrofa domestica*) crania were used. Penetrating trauma was inflicted to each pig's crania using both flared-tipped and parallel-tipped screwdrivers. The screwdriver shafts were 5mm in diameter and the shafts were attached to a drop rod to maintain consistent force for each trauma. The screwdriver was withdrawn after each wound was inflicted. The skull caps were removed and macerated. Tool mark analysis was conducted using a Leica® microscope. The following features were analyzed: (1) shape of penetrating wound; (2) length and width of penetrating wound; (3) wound margins (straight or irregular); (4) number

and type of radiating fracture (straight or curved) with respective angle; and, (5) number, location, and size of hinges. A Tree model was used to analyze the data. Preliminary results suggest that a distinction between flat-headed flared-tipped and parallel-tipped screwdriver penetrating wounds is possible. Length, margin and width yielded a classification system with 96% accuracy. Using radiating fractures, a distinction was possible using angle, location, and type which provided 80% accuracy in distinguishing flare-tipped from parallel-tipped screwdrivers. Using hinge location and size, a distinction was still achieved; however, only 66% of screwdrivers were correctly classified. Wound shape appears not to be efficient when distinguishing between the two screwdriver types.

In summary, this study suggests that tool mark analysis of screwdriver trauma to the cranium may provide evidence of screwdriver type, specifically, distinguishing between flared-tipped and parallel-tipped screwdrivers.

References:

1. Federal Bureau of Investigation, 2011. Murder victims, expanded homicide data Table 8. Available at: <http://www.fbi.gov/about-us/cjis/ucr/crime-in-the-u.s/2011/crime-in-the-u.s.-2011/tables/expanded-homicide-data-table-8> [Accessed 12/07/2013].
2. Shaw K, Chung J, Chung F, Tseng B, Pan C, Yang K, Yang C. A method for studying knife tool marks. *J Forensic Sci* 2011;56(4):967-71.
3. Freas L. Assessment of wear-related features of the kerf wall from saw marks in bone. *J Forensic Sci* 2010;55(6):1561-9.
4. Parmar K, Hainsworth S, Ruddy G. Quantification of forces required for stabbing with screwdrivers and other blunter instruments. *Int J Legal Med* 2012;126(1):43-53.

Screwdriver Trauma, Cranium, Tool Mark

H49 Examination of Saw Blade Teeth-per-Inch Measurements in Bone

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After attending this presentation, attendees will understand basic principles of saw mark analysis and several factors of bones and saws that may lead to variability of Teeth-per-Inch (TPI) measurements in bone.

This presentation will impact the forensic science community by demonstrating what needs to be considered in experimental research in saw mark analysis before attempting to provide error rates associated with quantifiable traits, such as measurements reflecting TPI of saw blades.

Tooth hop and tooth imprint are both measurable traits used in saw mark analysis to estimate the TPI of saw blade(s) in dismemberment cases. Tooth hop refers to striae across the kerf wall of a cut that show patterned waves. Tooth imprint refers to residual imprints from saw teeth in the kerf floor or the breakaway spur. For both traits, the distance from one wave's peak to another was measured as the Distance Between Teeth (DBT). Several hypotheses were considered in this research: tooth hop can accurately be measured to estimate TPI of a saw; tooth hop is more likely to occur where more cortical bone is present; tooth hop is less variable in denser bone; and a chisel-tooth blade is more variable than a crosscut blade.

This study utilized two saw blades of varying TPI and tooth type (a six TPI chisel-tooth and an eight TPI crosscut). Two species were also used (white-tailed deer long bones and human femora). Sample sizes of individual groups were as follows: 32 human crosscut; 32 human chisel-tooth; 62 deer crosscut; and, 55 deer chisel-tooth cuts. Only one side of each complete cut

was analyzed. All cuts were assessed for tooth hop and imprint, which were then measured and photographed for DBT under a stereomicroscope. Thirty DBT measurements were also randomly taken between consecutive teeth down each blade to compare the variability of DBT from the actual blades to the variability of DBT taken from the bones. Overall images of each complete cut were used to quantify the amount of cortical bone and medullary space present. Trabecular bone was combined with medullary space because it was considered uninformative. Cortical thickness was also estimated. Welch Two Sample *t*-tests were performed to check for significant differences ($p < 0.05$) between the DBT measurements of saws and species. Chi-squared tests compared observed and expected values for the presence/absence of tooth marks on the bones and the location of the marks on bone (wall or breakaway spur) between saws and species. Step-wise Discriminant Function Analysis (DFA) was used to compare groups with and without marks to estimate what variables were significantly different between the groups. Finally, multiple linear regressions were used to examine the correlations of the number of marks present to the cortical thickness, as well as other parameters, including the areas and perimeters of the cortical bone and medullary spaces.

Mean DBT measurements taken from the bones are not significantly different from mean DBT measurements taken directly from the saw blades. There are also no significant differences between species. However, the six and eight TPI blades are significantly different from each other. The chi-squared tests show that the two saw types left an equivalent amount of tooth marks on the two species. In terms of locations of tooth marks, marks are significantly present more on walls than on breakaway spurs ($p < 0.05$). In DFA, the classification rate of marked bones versus unmarked bones is 63%. Stepwise selection suggests cortical thickness as the main discriminator between the two groups. However, when using multiple linear regressions, no significant correlation exists between the numbers of tooth marks in bone to the cortical thickness. These values become slightly more significant after the removal of dummy variables (bones that have no marks); nevertheless, their *p*-values do not fall below 0.05. Similarly, no correlations are found between the number of tooth marks and the other listed parameters.

In summary, all aforementioned hypotheses are confirmed. However, the variability found within the human bone measurements needs to be studied further in order to understand how DBT (and thus TPI) measurements are being affected. The more we understand about a material and what is cutting that material, the better we can estimate TPI measurements of saw blades used in dismemberment cases.

Dismemberment, Tooth Hop, Teeth Per Inch

H50 The Effect of Angle at Impact and Magnification Level on Striation Pattern Recognition

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After attending this presentation, attendees will understand the basic principles of sharp force trauma analysis, class characteristics associated with serrated and non-serrated blades, and how angle at impact and magnification level used during analysis influence the accuracy of blade type determinations.

This presentation will impact the forensic science community by introducing a methodology grounded in quantitative

observation for the assessment of knife serration patterns which are left on cut surfaces.

Six knives were utilized in this study to create experimental cut marks in porcine costal cartilage (*Sus scrofa*), including four serrated knives and two non-serrated knives. Two rounds of cuts were made with the goal of ascertaining a baseline accuracy level. Angle at impact was not specified and magnification level was not controlled during the examination for these rounds. Once the cut marks were produced, the specimens were preserved in a 10% formalin solution and then evaluated using white light microscopy. The cut surface of each specimen was examined for the presence of patterned striations and a determination of "serrated" or "non-serrated" was made. To examine how angle at impact influences the accuracy of blade determinations, a third round of cuts were made where each knife impacted cartilage specimens at five different angles and the specimens were examined at five different magnification levels, which resulted in a total of 150 observations (six knives x five angles x five observations). The cut surfaces of the specimens produced during the third round were examined for the presence of patterned striations as well, but magnification level was strictly controlled. All of the specimens were examined under each magnification level before being re-examined under each subsequent magnification level. The samples were also re-coded before progressing to the next magnification level, which ensured that blade determinations were blind. This portion of the study was included in order to ensure the reliability of the results, while also illustrating the impact of magnification level on the recognition of striation patterns. In total, there were 210 observations included in this study (60 observations in rounds 1 and 2; 150 observations in round 3).

Blade-type determinations were made with 100% accuracy for all specimens included in this study, which demonstrates that serrated blades are distinguishable from non-serrated blades. Serrated blades produce distinct, patterned striations as they cut through cartilage, whereas non-serrated blades produce fine, unpatterned striations or no visible striations at all. This research also demonstrates that angle at impact does not affect the overall accuracy of blade-type determinations. However, the distance between striations on the cut surface does vary based on angle at impact, with cartilage specimens impacted at 15° and 25° angles displaying more striations than cartilage specimens impacted at 75° and 90° angles. This means that the distance between striations on the cut surface of cartilage specimens impacted at 15° or 25° angles is more challenging to measure, but the high number of striations allows for patterns to be more readily identified, thus leading to more confident blade type determinations.

The accuracy of blade-type determinations was also not influenced by magnification level used during examination. The use of lower magnification levels, such as 10x or 20x, is sufficient to make a blade type determination of serrated or non-serrated. These findings suggest that sharp force trauma analysts should select magnification levels at their own discretion and should not feel bound to use higher magnification levels in their examinations. In addition, these findings indicate that standard light microscopy techniques, which are readily available to the majority of forensic scientists, are sufficient in making accurate blade-type determinations and measuring striation patterns in cartilage.

Sharp Force Trauma, Class Characteristics, Striation Pattern Recognition

H51 The Utility of GIS in the Spatial Analysis of Saw Cut Marks on Bone

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After attending this presentation, attendees will have an appreciation for how Geographic Information Systems (GIS) can be used in a nontraditional manner in the identification of saw cut patterns on bone.

This presentation will impact the forensic science community by adding a new tool to the study and interpretation of saw cut marks on bone.

GIS have been used extensively for storing and analyzing raster (image) and vector (point, line, and polygon) data covering large spatial extents. Because of its ability to analyze spatial data and patterns, GIS have more recently been applied to micro-scale features of bones.

Kerf wall striations, while highly visible, have proven for the most part to be ineffective in defining saw class. GIS appears well-suited to recognize saw cut striations as spatial patterns. To test the utility of GIS, a series of test cuts were made using long bones of *Odocoileus virginianus*. In this initial study two saws were chosen with reasonably different class characteristics: 11 Teeth Per Inch (TPI) cross-cut pruning saw (blade 1); and, 32 TPI wavy set hacksaw (blade 2). Using a Keyence® digital microscope the kerf wall striations were digitally recorded in a jpeg format. The resulting jpeg images were processed to remove the background to eliminate background bleeding onto the striation surface.

Using ArcMap™ 10.1, each jpeg image was separated into three color bands (blue, green, and red). Band 3, the blue band, was not used. To analyze the variation within each image, an Isocluster Unsupervised Classification in ArcGIS 10.1 was applied. This analysis determined the maximum number of statistically similar cluster classes based upon the image reflectance patterns. The more homogeneous the image (low texture, smooth surface), the fewer classes required to represent the spectral variation in the image. FRAGSTATS spatial pattern analysis software was used to examine the pattern and spatial distribution of the cluster classes and to compare patterns produced from different saw blades.

FRAGSTATS analyzed the striation patterns using two values: Patch Richness and the Interspersion-Juxtaposition Index (IJI). Patch Richness (number of cluster classes) provides an indicator of the overall variation in reflectance within a given image. Images having a smoother and more homogenous surface will tend to result in fewer classes (lower richness), whereas those having a more textured surface will tend to produce a higher number of clusters. Eight trials using saw blade 1 (11 TPI cross-cut) produced an average of 25 classes (range 14-39), whereas five trials for blade 2 (32 TPI wavy cut) produced an average of 8 classes (range 7-9). This indicates a greater degree of variation in reflectance among the red, blue, and green bands for images from blade 1 and thus more surface variation (and shadowing) texture. Interspersion-Juxtaposition Index (%) refers to the spatial intermixing of different patch types and increases in value as patches tend to be more evenly interspersed. Potential values range from 0-100% with 100 indicating that all patch types are equally adjacent to all other patch types (maximum interspersion). This is an indicator of the total edge increasing as patch shapes become more irregular (as the image contains more edge relative to interior of cluster classes (patches). A single square patch (homogenous image) would produce a value of 1. Saw blade 1 had an average IJI of 68% while blade 2 produced an average of 77%, indicating that blade 2 produced fewer and more homogenous classes, each occupying a larger proportion of the image area and thus likely to be adjacent to different class types.

This initial study has demonstrated that GIS can be used in the pattern analysis of saw cut striations. Further analysis is needed to establish the Patch Richness and Interspersion-Juxtaposition Index for additional saw classes and formulate a methodology for

testing an unknown striation pattern against known patterns.

GIS, Saw, Cutmarks

H52 Postmortem Damage to Skeletal Elements Due to the Extreme Weather Conditions of Hailstorms

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After attending this presentation, attendees will be familiar with a detailed description and patterning of the postmortem damage to the skeleton caused by extreme weather conditions (e.g., hail). The new information will allow anthropologists to improve the accuracy and specificity of postmortem defects observed on skeletal remains.

This presentation will impact the forensic science community by introducing unexpected postmortem fractures resulting from a weather-related phenomenon. Attendees will be provided a reference for a new interpretation of specific postmortem damage that might have been overlooked. In the future, the attendees will be aware of hailstones as a possible explanation of previously unexplained postmortem damage.

No known documentation of hailstorm damage to the human skeleton exists in the forensic literature. Clinical data indicates that hailstones can cause severe bruises and/or fractures on living people similar to that which is seen in blunt force trauma. Hailstones, which are irregular clumps of ice typically measuring from 5mm to 15cm in diameter, produce damage with a small impact area but with a high velocity. Thus, damage to the human body, including bone, can be severe.

On April 27, 2011, a band of strong storms caused record-setting wind and hail damage across East Tennessee. The Anthropology Research Facility (ARF — an outdoor laboratory for the study of human decomposition) in Knoxville, Tennessee, was hit by a downdraft that brought high winds and a severe hailstorm, causing the uprooting of trees, the topping and felling of trees, significant structural damage to the fencing, and widespread hail-related damage from hailstones observed to be approximately the size of chicken eggs. All body donations at the ARF at the time of the storm, were surveyed to determine the extent of that storm-related damage. Hailstone damage was noted for several of the human remains residing on the surface, which was expected, as it has been estimated that a hailstone with 8cm diameter can fall as fast as 171km/hour.¹

This study examined 19 skeletons, which have now been accessioned into the Bass Donated Skeletal Collection and were at the ARF on April 27, 2011. These individuals had been placed at the ARF mostly in a prone position between May 2010 and February 2011 with a postmortem interval ranging from nearly three months to twelve months. The locations varied throughout the ARF. Nine individuals are males and ten are females with age-at-death ranging from 42 to 82 years. All skeletons were thoroughly examined and any damage was documented.

The most commonly affected elements were thin bones like scapulae and ilia. Fourteen out of nineteen had at least one scapula fractured. Seven ilia and one ischium showed postmortem damage that was likely caused by hail. Three individuals had one of their zygomatic bones broken off as well as the maxilla damaged. One of these three exhibited a circular hole in the temporal bone. One cranium had major portions of the left temporal and parietal broken into multiple pieces. Interestingly, some lower limb long

bones exhibited postmortem fractures attributable to hailstone damage. Also, five individuals had postmortem fractures of the spinous processes of thoracic or lumbar vertebrae. Multiple individuals demonstrated postmortem fractures of the ribs, but the damage observed cannot be clearly linked to hail-related damage due to the general fragility of the ribs.

The damage caused by hail can be characteristics as circular or elliptical depression fractures of various sizes. Most of the fractures on the scapulae are complete circular depression fractures with the central piece missing. The fractures in the femora, tibia, fibula, and most of the ilia are incomplete, ovoid-shaped, depressed fractures. On narrow surfaces, like the ribs and spinous processes, the small surface area precludes determination of the shape of the impact site. However, unlike the damage to the ribs, the damage observed on the spinous processes is likely the result of hail.

Hail damage could be confused with other types of postmortem damage but the circular shape of the impact area and complete perforation of the bone indicate a small ovoid-shaped object with high velocity hitting the surface. When unusual postmortem damage is observed in remains recovered from the outdoor context, hailstone damage should be considered.

Reference:

1. <http://www.nssl.noaa.gov/education/svrwx101/hail/>

Taphonomy, Blunt Force Trauma, Hail

H53 A Forensic Pathology Tool to Predict Pediatric Skull Fractures — Part 4: Interface Effects on Head Drops

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The goal of this presentation is to inform attendees about research on the influence of impact interface on fracture patterns in controlled head-drop experiments onto the parietal bone with a subadult porcine (*Sus scrofa*) model.

This presentation will impact the forensic science community by comparing controlled head drops onto rigid and compliant interfaces.

In forensic investigations, distinguishing accidental from inflicted injury based on skull fracture alone is challenging as both cases may produce similar patterns. From this, the research team identified the need for a deeper understanding of the fracture mechanics of the skull through controlled simulation of traumatic head injuries.

Recent studies have demonstrated the utility of an infant porcine model in simulating impacts to the developing human skull. It has been shown that increasing the impact energy applied to entrapped heads increases the number and length of cranial fractures.^{1,2} The current study examines the effect of interface compliance on skull fractures from head drops in the porcine model.

The hypotheses of this study were twofold: first, head drops onto a rigid interface would produce more fracturing than drops onto a compliant interface; and second, fracture patterns produced in high-energy head drops onto a compliant interface would be similar to those produced by lower-energy drops onto a rigid interface. To generate a single impact onto the center of the right parietal bone, a custom drop tower was developed with an adjustable height. Specimen heads of a given age could be dropped with a consistent orientation onto various surfaces at the

same impact energy. Pigs that had died of natural causes between 3 and 19 days of age (n=87) were collected and frozen within 12 hours of death for this study.

The study was performed with controlled head drops onto a rigid surface and two carpeted interfaces. The rigid surface was a half-inch thick aluminum disk (n=32). "Carpet 1" interface covered the rigid surface with a thin, commercial-grade carpet (n=23) and "carpet 2" interface covered the surface with a thick polyester carpet with an underlayment (n=32). After impact, the crania were inspected for diastatic fractures, the soft tissue was removed, and the dry cranial bones were reassembled. Fractures were recorded on a standard diagram and the location and length of each was recorded to the nearest millimeter.

Total fracture length from head drops onto the rigid surface was significantly higher than both carpet 1 ($p=0.002$) and carpet 2 ($p<0.001$). Carpet 1 also produced a significantly higher total fracture length than carpet 2 ($p=0.009$).

Carpet 2 data was compared to previously published low-energy drops onto a rigid interface (1). Total fracture length ($p=.259$) and total number of fractures ($p=.204$) showed no significant difference. However, low-energy rigid drops produced significantly more diastatic fractures ($p<.001$) and fractures that crossed through a suture ($p=.049$). The low-energy rigid drop tests also tended to produce more fractures in bones other than the impacted bone ($p=.057$).

In summary, this study showed that a head dropped onto a rigid interface would produce significantly more total fracturing than one dropped onto a compliant interface. Energy absorbed through deformation of the compliant interface reduces the impact energy imparted to the skull to lessen cranial fracturing. Furthermore, while the degree of fracturing was similar for high-energy drops onto a compliant interface as low-energy drops onto a rigid interface, the fracture patterns were different, so that these impact scenarios would be distinguishable post-trauma. These results demonstrate that impact interface may be a significant factor in the interpretation of injury causation that involves cranial fractures in the pediatric trauma victim.

This project was supported by the National Institute of Justice, Office of Justice Programs, United States Department of Justice. The opinions, findings, and conclusions or recommendations expressed in this presentation are those of this study and do not necessarily reflect the views of the Department of Justice.

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2. Powell BJ, Passalacqua NV, Baumer TG, Fenton TW, Haut RC. Fracture patterns on the infant porcine skull following severe blunt impact. *J Forensic Sci* 2012;57(2):312-17.

Cranial Fractures, Biomechanics, Child Abuse

H54 Pediatric Cranial Fracture Patterns in Cases of Head Trauma Resulting in Death

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After attending this presentation, attendees will understand cranial fracture pattern differences between pediatric homicides and severe accidents (falls from heights and crushing injuries, such as a flat screen TV falling on an infant). The distribution of fracture types, the common fracture site, the number of bones fractured in each case, the average number of fractures, and the frequency of cases where fractures crossed sutures will be presented.

This presentation will impact the forensic science community by providing medicolegal death investigation experts (such as forensic pathologists, coroners, forensic anthropologists, and medical examiner investigators), as well as child protective services professionals, very recent data that may help in the interpretation of potential causes for pediatric cranial trauma.

In medicolegal death investigations, current techniques for interpreting pediatric cranial trauma are of questionable reliability due to a lack of baseline data that matches pediatric cranial fracture patterns with known impact scenarios. The current research addresses this significant gap in best practice through a multidisciplinary effort to collect data on human pediatric deaths involving blunt force cranial fracture and known impact scenarios from current forensic case files at medical examiner offices across the country with an ultimate goal of establishing a national website of such data (The Pediatric Cranial Fracture Registry).

A total of 66 head trauma cases involving children (37 boys and 29 girls) with clear autopsy photographs and/or diagrams of sustained cranial fractures were analyzed. In 55 cases, the manner of death was homicide due to blunt force trauma and/or cranio-cerebral injury. The mean age was 11 months. In 11 cases the manner of death was accident (six cases were crushing accidents, such as a flat screen TV falling on an infant; five cases were falls from heights). The mean age of this group was 2 years.

The distribution of fracture types for homicides was: 73% multiple or complex; 22% single linear; and, 5% comminuted. In accidental cases, 100% of the fractures were multiple or complex. The frequency of accident cases with diastatic fractures was 91%, whereas only 43% of homicides displayed diastatic fractures. Basilar fractures were observed in 91% of the accident cases but in only 2% of homicides. In 91% of accident cases fractures were crossed sutures compared with only 38% of homicide cases.

The number of cranial bones fractured in each case can be a measure of injury severity. In accidents 0% of cases displayed one fractured bone, 27% two fractured bones, 9% three fractured bones, 37% four fractured bones, and 27% five fractured bones. In contrast, homicides displayed one fractured bone in 24% of cases, 29% two fractured bones, 29% three fractured bones, 13% four fractured bones, and 5% five fractured bones.

Analysis of the fracture sites in accidents indicated the parietal bone was the most commonly fractured bone (81% of cases), followed by the frontal (73%), temporal (64%), sphenoid (64%), and the occipital (45%). In homicide cases, the parietal bone was again the most commonly fractured bone in 82% of cases, followed by the occipital bone (58% of cases), the frontal bone (25%), the temporal bone (22%), and finally, the sphenoid (5%).

The percentage of accident cases by fracture number (including segments of a complex fracture) was: 27% of cases displayed one fracture; 9% two fractures; 0% three fractures; 0% four fractures; 0% five fractures; 0% six fractures; 0% seven fractures; 18% eight fractures; 9% nine fractures; and, 37% ten or more fractures. In homicides, 22% of cases displayed one fracture, 13% two fractures, 15% three fractures, 9% four fractures, 13% five fractures, 4% six fractures, 4% seven fractures, 4% eight fractures, 7% nine fractures, and 9% ten or more fractures.

This study showed that accidental pediatric deaths involving severe falls and crushing accidents involved more:

basilar fractures, diastatic fractures, multiple or complex fractures, fractures crossing sutures, fractures in the frontal, temporal and sphenoid bones, total fractures, and bones fractured per case.

This project was supported by Award No. 2011-DN-BX-K540, awarded by the National Institute of Justice, Office of Justice Programs, United States Department of Justice. The opinions, findings, and conclusions or recommendations expressed in this presentation are those of this study and do not necessarily reflect the views of the Department of Justice.

Pediatric, Fracture Patterns, Blunt Force Trauma

H55 Trauma Patterns of Accidental and Intentional Injury

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After attending this presentation, attendees will be informed about current research and methods utilized in characterizing fractures that result from accidental and intentional trauma. Attendees will understand how this analysis will take patterns of traumatic injury, demographic characteristics, and features of the injuries into account in an effort to discern patterns between the two causes of injury.

This presentation will impact the forensic science community by demonstrating how fracture patterns within a contemporary skeletal collection conform to known research regarding the patterns expected for intentional and accidental injury.

The ability to determine the cause of skeletal trauma — that is, a bodily injury produced by blunt, sharp, or ballistic forces — is critical in assessing the manner of death as homicide, accident, suicide, natural, or unknown. Current scholarship demonstrates that injuries resulting from accidents tend to be more widely distributed throughout the skeleton, while injuries resulting from intentional causes have a tendency to be more localized. However, researchers have not yet adequately addressed if there is a unique, identifiable pattern inherent to the injuries that result from different causes (i.e., an accident or intentional injury) or what the statistical specificity and sensitivity of these patterns is likely to be. The purpose of this study was to examine the patterns of injury between known accidental and intentional trauma cases while analyzing fracture features, and the location of injuries in individuals of varying ages, sexes, and ancestries. The William M. Bass Donated Skeletal Collection was utilized for this study and provides a sample of modern individuals (N=66). Information was collected in three areas, demographics, fracture features, and injury features. First, demographic characteristics of individuals (i.e. age, sex, and ancestry) will be recorded for each skeleton. If sex, age, or ancestry could not be determined, then they were labeled as unknown in that category. Second, fracture features (e.g., type, size, etc.), and fracture location (i.e. bone, side, specific location) were assessed via macroscopic examination of skeletal material and radiographs. This data will allow a further elucidation of what kinds of injuries occur from different causes, as well as what areas of the bone may be biomechanically more susceptible to certain injuries.

Statistical analysis of this sample shows that there are differences between the accidental and intentional trauma types regarding some of the features in question. Chi-square analyses have shown that intentional injuries in this sample tend to be less distal than expected and have more ballistic and radiating fracture types than expected ($p < 0.05$). Accidental injuries tend to be overall more varied by location and tend to have less ballistic fracture types and more crush injuries than expected ($p < 0.05$). Concerning the location of injuries, intentional trauma had fewer injuries to the right

side than expected and more injuries than expected for trauma that involved both sides. Accidental trauma had more left sided injuries than expected and fewer injuries than expected in cases where both sides were affected. Males in this sample exhibited more intentional injuries than expected, while females had less intentional injuries than expected and more accidents than expected.

These findings show that there are statistically significant differences between accidental and intentional trauma types for the selected sample as concerns demographic and fracture characteristics. The patterns conform in some respects to established literature; for example, the intentional injuries tended to be less distal than expected. This analysis is preliminary, however, and highlights the need for further research to fully define these patterns and assess their utility in different regions.

Trauma, Fractures, Accident

H56 Pedestrian-Motor Vehicle Accident Trauma: Pattern Interpretation and the Promotion of a Multidisciplinary Approach

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After attending this presentation, attendees will have a better understanding of the importance of fracture pattern interpretation in the reconstruction of a pedestrian-motor vehicle accident. The main goal of this research is to illustrate the importance of a multidisciplinary approach to trauma analysis in pedestrian-motor vehicle accidents in a medical examiner/coroner setting.

This presentation will impact the forensic science community by illustrating the anthropological contribution to accident scene reconstruction and fracture pattern interpretations.

Pedestrian-motor vehicle accidents are documented as having the highest mortality rates of all motor vehicle traumas and this type of accident is one of the primary causes of blunt force trauma in the forensic context.^{1,2} Additionally, in the case of a hit-and-run accident, anthropological assessment of the fracture patterns may be crucial to the interpretation of the traumatic incident as witness accounts are not always reliable or consistent. The contributions of a forensic anthropologist to accident reconstruction can include, but are not limited to, analysis of the direction of impact, minimum number of impacts, distinguishing between direct and indirect trauma, and interpretation of the victim's stance at the time of impact (i.e., weight bearing vs. non-weight bearing elements).

Previous studies have assessed patterns of traumatic injury observed in pedestrian-motor vehicle victims and have suggested two patterns of skeletal injury demonstrative of a typical pedestrian motor vehicle trauma: the "fatal triad" and the "ipsilateral dyad."¹ The "fatal triad," as coined by Farley in 1965, consisted of simultaneous fracturing of the skull, pelvis, and extremities.³ The "fatal triad" was subsequently modified by Waddell and Drucker to delimit extremity fractures to the region around the knee.⁴ A more recent study by Brainard *et al.* attempted to confirm the work of Waddell and Drucker and found that instead of the "fatal triad," a large percentage of pedestrians struck by motor vehicles exhibited an "ipsilateral dyad" of traumatic injury consisting of fractures of both the upper and lower extremity on the same side of the victim.¹ Additionally, Brainard *et al.* found an association between femoral fractures and corresponding pelvic fractures.¹

The current study analyzed 28 cases of pedestrians struck by motor vehicles between the years 2003 and 2013. All

cases were drawn from the Maricopa County Forensic Science Center in Phoenix, Arizona. Victims ranged in age from 16 years to 84 years old at the time of death. The study included both males and females; however, 26 of the 28 victims in this study were male (93%). Skeletal analysis consisted of an examination of the following elements: femora, tibiae, fibulae, and patella. In addition to the skeletal analysis, data were also drawn from autopsy radiographs and pathology reports. Unlike the previous studies conducted by Farley and Brainard *et al.*, this study consisted of only fatal pedestrian-motor vehicle accidents.^{1,3} Therefore, this study represents only a small subset of pedestrian-motor vehicle traumas, being that all individuals included in this study died as a result of their injuries.

In the present study, the most frequently fractured elements were the pelvis and the ribs, which were both fractured in 18 of the 28 victims (64%), followed by the fibulae (63%), skull (61%), tibiae (59%), vertebrae (46%), and femora in 18% of the cases. Noted the presence of Waddell and Drucker's "fatal triad" (head, pelvis, knee) in four of the 28 cases (14%) and the "ipsilateral dyad" in six of the 28 cases (21%).⁴ However, when lower extremity injuries were incorporated into the category of knee injuries the "fatal triad" was observed in 10 of the 28 cases (36%). Supporting Brainard *et al.*'s findings, in 90% of the cases involving femoral fractures, associated pelvic fractures were also noted.¹

In conclusion, 17 of the 26 (65%) cases (with accident reports) were hit-and-runs where the accident circumstances were either absent or conflicting. When the forensic anthropological analysis was incorporated into the forensic pathologist's report, direction of impact was possible to infer in 82% (23/28) of the cases based on fracture pattern interpretation. This finding provides justification for the promotion of a multi-disciplinary approach in the interpretation of pedestrian-motor vehicle accidents.

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Pedestrian-MVA Accidents, Fracture Interpretation, Trauma Analysis

H57 Decomposition in Western Montana: Defining Postmortem Changes in the Fall, Winter, and Spring

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After attending this presentation, attendees will better understand the processes of decomposition in western Montana for remains deposited outdoors in the fall, winter, and spring and will have gained knowledge of better Postmortem Interval (PMI) estimation in an environment with extended periods of cold ambient temperatures.

This presentation will impact the forensic science community by providing a baseline dataset for the rate and sequence of decomposition in western Montana allowing for more accurate estimations of PMI by forensic anthropologists.

Little is known about how remains decompose in western Montana; however, preliminary decomposition studies indicate that the decay processes do not follow expected rates found in other geographic locations. Western Montana's climate is unique; it can be extreme and fluctuate rapidly, even hourly. The arid environment that is found throughout this region provides four distinct seasons that differ in their humidity and temperature. Understanding how the unpredictable environment affects human remains is crucial for forensic anthropologists in estimating PMI. Without an expected pre-established baseline dataset, estimating Time Since Death (TSD) can be difficult, especially in an environment like western Montana, where approximately six months of the year ambient temperatures are around and below freezing. This leads to the important research question; do remains deposited during the winter months show specific indicators that can be used to better obtain TSD.

This study utilized three adult pigs (*Sus scrofa*) ranging in weight from 68-90kg as human proxies which were acquired from a local Montana Rancher. Specimens were deposited in two separate locations, both on private ranches: fall (October 1, 2011) and winter (December 2, 2011) were deposited in the Missoula Valley and spring (May 26, 2012) in the Bitterroot Valley, approximately 20 miles south of Missoula. Each specimen was deposited on the surface and enclosed in a 1.8m x 3.0m x 1.8m prefabricated dog kennel which was protected by electric fencing to deter scavenging and disruption by ranch animals. Data was collected using procedures established for western Montana by Gonder and analyzed by comparing the following variables: ambient temperature; relative humidity; weather patterns; internal temperature; external temperature; bloat; odor; color; entomological activity; and other visual observations.¹ For the purpose of this study, the Galloway *et al.* method was altered to more accurately reflect the decompositional changes of the specimens. Four stages were used: (1) fresh; (2) early decomposition; (3) advanced decomposition; and, (4) mummification.²

Results of this study indicate that the overall patterns of decomposition are similar compared to studies conducted in other geographic locations; however, unscavenged remains ultimately result in mummification and not skeletonization. The rate of decay for the three specimens differed from one another and deviated from standard expected results. Due to Montana's cold winter temperatures, the remains deposited in the fall and winter decomposed at slower rates and both experienced stasis in the decay process as a consequence of ambient temperatures below 4.4°C. The remains deposited in the fall stayed in the fresh stage for 1 day, early stage for 26 days, advanced stage for 17 days, stasis for 116 days, advanced stage for 125 days, then mummification for 80 days until the conclusion of the study.² The remains deposited in the winter stayed in the fresh stage for 7 days, early stage for 19 days, stasis for 61 days, early stage for 75 days, advanced stage for 69 days, and mummification for 134 days until the conclusion of the study. The remains deposited in the spring stayed in the fresh stage for 1 day, early stage for 14 days, advanced stage for 14 days, and mummification for 248 days until the conclusion of the study.³ Results of this study also indicate that there are specific identifiers that are useful in estimating PMI in cold weather environments.

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Decomposition, Postmortem Interval, Montana

H58 How Black-Billed Magpie Scavenging Can Skew Postmortem Interval Estimates in Western Montana

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After attending this presentation, attendees will understand how estimates of Postmortem Interval (PMI), utilized from several current methods, are not appropriate for remains that have been scavenged by facultative black-billed magpies in western Montana and, therefore, should not be used.

This presentation will impact the forensic science community by challenging forensic anthropologists to elevate their knowledge of micro-environmental and regional taphonomic patterns and paradigms, especially those that incorporate the estimation of postmortem intervals.

The PMI in Montana is difficult to establish, as most of the data sets and methods developed to estimate the PMI have been developed for only a few bioclimatic zones. Bodies found deceased in Texas, Indiana, or Florida, for example, will decompose at a different rate than bodies in Montana. Applying the same PMI methods to all climatic zones will likely result in inaccurate estimates of PMI. Likewise, the condition or environment in which the body is discovered (e.g., water submersion, mummified, hanging, burial, burned) can also affect the PMI estimation.

In order to examine the decomposition processes in western Montana, three pig (*Sus scrofa*) cadavers (SS1, SS2, and SSC, the control cadaver) were placed in a research facility on a ranch just outside of the Missoula, Montana, city limits from August 1, 2011 through August 20, 2102, in two separate micro-climates (full sun versus full shade). Data from the decomposition process, climatic variables, entomological activity and Black-billed Magpie scavenging were observed for over one year. Researchers involved in determining stages of decomposition for estimating PMI need to be cognizant of the taphonomic impacts avian scavengers can have on the decomposition process. The research goal was to determine what variables had the biggest effect on the decomposition specimens in this study. All three cadavers advanced from the fresh stage of decomposition to the advanced stage of decomposition within nine days, at which time desiccation in the form of mummification took place. Full skeletonization did not occur, but after 120 days at least 50% of the skeletal remains were visible on SS1; after 250 days at least 50% of the skeletal remains were visible on SS2; and, at no point did SSC experience skeletonization. The Black-billed Magpies were almost wholly responsible for the skeletonization process.

The hypothesis posited is predicated on the supposition that cadavers that are placed outdoors in the hot, semi-arid environment of western Montana, in August will mummify rather than advance to the skeletonization stage without the aid of avian or terrestrial scavengers.

Several studies have been conducted on obligate avian scavengers (vultures), but there has never been a study on how the facultative Black-billed Magpie scavengers affect decomposition and, consequently, skew the PMI. In this study, it was impossible to prevent avian and small rodent scavenging without making alterations to the enclosures. As this was a longitudinal study looking for the variables that effect decomposition, scavenging

was allowed to take place on SS1 and SS2, but prevented from accessing SSC, which was used as the control cadaver. The considerable influence of Black-billed Magpie scavenging on the decomposition process in this research study demonstrates that future studies need to consider what scavenging species are in the environment, both spatially and temporally, and where a human body may be found. Avian and terrestrial scavenging accelerate decomposition to such an extent that the PMI estimates can be thrown off by days, months, or even years. For the most accurate estimations of PMI, all naturally occurring variables such as scavenging need to be included in decomposition studies.

The greatest observation from this research is that there are too many stochastic variables to consider in each bioclimatic zone to attempt to make a broad statement about taphonomic "universals." In closing, the only way to accurately estimate a PMI is to incorporate all variables that effect decomposition. It is our responsibility as forensic scientists to continually look for better answers, to toss out outdated methods, and open our minds to new methods.

Black-Billed Magpie Scavenging, Postmortem Interval, Decomposition

H59 Is Carpal Tunnel Syndrome Detectable in the Skeleton?

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After attending this presentation, attendees will understand how carpal tunnel syndrome manifests in the skeleton and how this can be used in building the biological profile.

This presentation will impact the forensic science community by providing forensic anthropologists with knowledge as to how carpal tunnel syndrome presents in skeletons and how this is related to obesity, which has the potential to increase the precision and accuracy of observations during forensic assessments.

Skeletal pathology accompanies obesity, including Diffuse Idiopathic Skeletal Hyperostosis (DISH) and Osteoarthritis (OA). In order to define a signature of obesity in the skeleton, it is important to assess how other pathological conditions may manifest in the skeleton and combine this knowledge with that of the frequency of DISH and OA. Carpal Tunnel Syndrome (CTS), which affects the hand, wrist, and forearm, has been linked with obesity in the clinical literature. Treatment of CTS includes changing the hand, wrist, and forearm movements of CTS patients and/or surgery to increase the size of the carpal tunnel to accommodate the median nerve. The changes to muscle movements (increasing/decreasing) should lead differential expressions of Musculoskeletal Stress Markers (MSM), or entheses, in the hand, wrist, and forearm.

In order to test the hypothesis that enthesal expression is modified in CTS patients, donors with documented CTS in the W. M. Bass Donated Skeletal Collection were examined. These individuals had body masses ranging from normal to obese. MSM development in the hand, wrist, and forearm for muscles involved in flexion/extension and supination/pronation (the movements modified in CTS patients) were assessed in each skeleton. However, muscular development for the wrist and hand muscles were negligible, and thus dropped from the study. No differences in muscular expression among CTS and non-CTS patients were evident using a chi-square test. This is not surprising, as MSM have come under recent scrutiny for their inability to accurately distinguish between activity levels and body weight. Interestingly, while recording entheses for markers of rusticity and stress, the presence of OA in the hand and wrist areas was noted in 66.7% of

the CTS patients.

In summary, detecting CTS in the skeleton is not straightforward and cannot be detected by muscular development via entheses. Further, a relationship between CTS and OA in the hand and wrist may exist, warranting additional exploration. However, this relationship is not strong enough to be diagnostic of CTS, alone. Thus, the forensic practitioner should consider looking for the suite of pathological conditions associated with obesity during the construction of a biological profile, especially if attempting to assess body mass from skeletal remains. If a suite of detectable conditions and/or diseases associated with obesity are present and considered in conjunction with other skeletal characteristics, the unknown individual may be obese. Also, the findings of OA in the hand/wrist of CTS patients may help to understand the etiology behind localized or isolated hand/wrist OA.

Identification, Biological Profile, Obesity

H60 Fracture Patterns in Postcranial Flat Bones Inflicted With High-Velocity Expanding Ammunition From Two Different Ranges

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After attending this presentation, attendees will gain insight into the fracture patterns created on postcranial flat bones shot by high-velocity expanding ammunition. The goal of this study is also to aid in better understanding the influence which shooting distance has on the pattern of injuries inflicted on postcranial skeletal elements.

This presentation will impact the forensic science community by furthering the understanding of gunshot injuries on flat bones caused by a civilian rifle. The information gained from this presentation will assist forensic anthropologists to infer the type of weapon and the approximate shooting distance based on the fracture patterns of postcranial flat bones. This is crucial in both civilian and military frameworks when reconstructing the circumstances of an individual's death.

The majority of the studies published on gunshot injuries have been focused almost exclusively on the cranium.¹⁻³ Very limited research regarding projectile trauma to the postcranial skeleton has been published and even less related to gunshot traumas inflicted by high velocity weapons.⁴⁻⁸ Most of this published research does not involve experimental studies; nor in many instances are the variables (bullet caliber, trajectory, etc.) involved in the production of the observed fractures actually known. Logic has been used to interpret these sequelae, but in the absence of experimentally derived data, the likelihood is equivocal.

An experimental study was carried out in both the TRACES facility and the forensic anthropology laboratory of the University of Central Lancashire using the domestic pig (*Sus scrofa*). Fifteen half-carcasses and eight heads were shot from a muzzle-to-target distance of 5m and fifteen half-carcasses and seven heads from a range of 20m, respectively, with a 0.243 caliber hunting rifle and as perpendicular as possible to the target. The scapulae, the ribs, and the mandibles were successively defleshed, macerated, and reconstructed. The resulting fracture patterns were classified according to the respective observations for each type of bone separately and compared for the two ranges. Radiographs of the mandibles and the scapulae were also obtained.

Preliminary results showed that the six mandibles which were directly hit by the bullet from the 20m distance presented a wide irregular entrance site on the target-ramus with a characteristic

radiating pattern and with no or minimal fracturing of the opposite ramus. On the contrary, the three mandibles shot from the 5m-range showed either a through-and-through pattern (i.e., entrance on the target-ramus and exit on the opposite ramus) or comminuted fracture of the opposite ramus. The higher amount of kinetic energy transferred to the bone tissue in the case of the 5m shooting range in combination with the composition of the mandibular bone tissue represent possible explanations for this difference in the pattern.⁹ The mandibles that were indirectly fractured (i.e., the maxilla was accidentally shot instead of the mandible) either from the 5m or the 20m distance showed similar pattern, comminuted in nature, implying that the expansion of the bullet inside the oral cavity caused this pattern irrespective of the distance of shooting.

The "Tree statistical analysis" performed for the scapulae revealed, with an accuracy of approximately 82%, that the presence of comminuted fractures was the main characteristic that differentiated the bones shot from the two distances. Additionally, the majority of the scapulae shot from the 5m distance presented exfoliation both toward the direction of fire and opposite to it; on the contrary, the scapulae shot from the 20m range showed exfoliation only on the surface toward the direction of fire. The higher degree of plastic deformation of the bone tissue caused by the missile in the case of the shorter distance of shooting may explain this observation. The analysis of the ribs revealed preliminarily that in order to determine the distance of firing, the specific location of the fracture (i.e., on the vertebral, middle or sternal third) plays an important role in the morphology of the resulting fracture patterns.

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Postcranial Bones, Fracture Patterns, High-Velocity Ammunition

H61 The Complete Forensic Anthropology Application for Smartphones and Tablets

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After attending this presentation, attendees will understand how to navigate a software application for smartphones and tablets which allows users to complete a biological profile of an unidentified individual. A trial of this software application program will be available following the presentation.

This presentation will impact the forensic science community by providing the necessary tools for a forensic anthropologist to complete a biological profile in the new age of technology. One of the most important steps in the scientific method is the retesting of ideas. Professionals will not only have access to the most used methods in the field but some of the newest ways will also be included so testing can be done. This library of reference material will be used to connect all professionals in the field together to share information and methods.

Is it time for identification technology to be in the palm of your hand? The answer is yes. In the modern world, information is no longer being shared through paper and pen but has been converted into a digital format. For example, the television signal has been changed from analog to digital and books are being downloaded and read on tablets. It is important that the different professional fields also keep up with these technological changes that occur in the world. Previous software only covers one area of the biological profile at a time. It is hoped that the combination of all the methods into one place will allow easier testing and completion of biological profiles in the field or morgue settings.

Forensic anthropologists compose a biological profile of skeletal remains of an individual in the hopes that identification can be made through the estimation of the age, sex, ancestry, stature, and any trauma that occurred to the individual. For each of these areas professionals have devised multiple methods using various skeletal areas to make estimations for identification. The goal of this app is to bring all the methods which are used to complete a biological profile and condense them into one easy to use location.

The program is divided into five sections: skeletal inventory; age assessment; stature assessment; sex determination; and ancestry determination. The skeletal inventory section allows for a quick evaluation of the bones that are present to take place. With a simple click, not only will the inventory become complete but a mapping of the fetal, subadult, or adult skeleton can be done.

The age assessment section examines methods for both subadult and adult calculations. Charts set up with one-touch selection allow for easier estimations to be done. The stature assessment employs the major methods used to calculate both whole and fragmented bones. Mathematical calculations are often tedious and with all the different units of measuring, can be confusing. This section allows the user to input the measurement taken for various methods and will present the estimated height of the unknown individual. The sex determination section allocates the methods for determination based on analysis of the pubic and cranial bones using an interactive guideline. In addition, new methods that analyze shapes, angles, and measurements of various long bones will be included.

The ancestry determination section uses image manipulation which contains standardized tools to allow for magnification and scrolling for better visualization of the skull reference to be compared to the case. Analysis of the data based on the individual is estimated and an output screen displays the final calculations depending on the method chosen.

In this presentation, a walkthrough and examples of possible uses of the app will take place. Members in the audience will be given a beta test of the app which will be available for six months after the presentation. The goal of the beta test will be to test the usefulness of the app with professionals in the field. Once a month, a questionnaire will be accessible to users of the app to hear what works and what needs to be improved. Once all the available data are gathered and all glitches are fixed, a full app will be launched. The ultimate goal of the application is to be not only a guide for professionals in the field, but also a teaching aid to students.

Forensic Science, Forensic Anthropology, Software Application

H62 A New 3D Landmark Reference Database for Sex and Ancestry Assessment in Human Skulls

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After attending this presentation, attendees will understand the principles on which advanced software tools for assessing an individual's biological sex and ancestry from a human skull are grounded, such as the importance of a large cranial reference database, the basics of geometric morphometrics (shape analysis) to comprehend advances in studies of craniofacial morphology, and their applicability for identifying skeletal remains.

This presentation will impact the forensic science community by presenting a collaborative project aimed to advance methodological and computer-assisted techniques currently available in the field of forensic anthropology and to facilitate and substantiate craniofacial examinations.

Portable Three-Dimensional (3D) devices (e.g., MicroScribe®, Polhemus® digitizers) have enabled researchers to rapidly acquire coordinate landmark data, are relatively user-friendly, and are accurate. If the use of data acquisition hardware is combined with methods of geometric morphometrics, they will augment classification rates for biological sex and ancestral affinity, in particular if based on craniofacial morphology. Additionally, new developments in 3D technologies have made it possible to facilitate medicolegal examinations in unidentifiable remains using non-invasive and non-contact approaches such as computed tomography and other more affordable 3D scanners that have modified the manner in which traditional osteometrics are executed. Virtual 3D data can be easily transported and shared between laboratories; they represent an unlimited source of morphological data and provide a real-time access for re-examination of physical evidence. Similar to direct digitization of skeletal remains, collecting 3D coordinates has been shown to be advantageous when identifying virtual skeletons than more traditional methods.

However, processing 3D landmark data involves a large learning curve, particularly in the area of theoretical background requiring computer-assisted shape analysis algorithms, and multivariate statistics, and integrating geometric morphometrics into an everyday routine has been challenging. Recently, in order to overcome these challenges the software programs 3D-ID developed by Slice and Ross and COLIPR developed by Urbanová and Králík were created. They both use a landmark-based approach combined with discriminant and canonical variates analysis in order to provide the best classification for an unknown individual. However, the performance of the algorithms and a broader applicability are mainly dependent upon the representativeness of the incorporated reference database. While 3D-ID includes eight groups with a minor coverage of European populations, COLIPR was developed to be used primarily in a context of European ancestry groups.

A new reference database has emerged from a collaborative project between forensic scientists and both software applications, which has given rise to a comprehensive cranial dataset of approximately 2,400 specimens with 16 distinct groups (Native Africans, Asians, Caribbeans, central Europeans, eastern Europeans, southeastern Europeans, southern Europeans, southwestern Europeans, Mesoamericans, African Americans, European Americans, Hispanic Americans, Afro-Brazilians, Japanese Brazilians, European Brazilians, Peruvians) based on their ancestry and geographical origin (twice as many if biological sex is included). Combined with additional methodological improvements in classification algorithms and online availability,

the new reference database will extend diagnostic power and applicability to a broader range of skeletal cases. For instance, an overall rate of biological sex estimation computed on 21 landmarks and the total number of incorporated groups reaches 81% of correctly classified cases if only shape variables are included and 87% of correctly classified cases if shape and size variables are combined. However, if input coordinates are adjusted according to population-specific cranial morphology and patterns of sexual dimorphism, the classification rate exceeds 95% of correctly classified cases.

Sex and Ancestry Assessment, Reference Database, Software Tools

H63 The Analysis and Variance of Strontium Isotopes in a Forensic Context: Determination of Geolocation Reliability for a Modern New England Population

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After attending this presentation, attendees will gain an understanding of how strontium isotopes in the environment are reflected in human enamel and bone and how its presence can be used to assist in the geolocation of unidentified human remains.

This presentation will impact the forensic science community by providing data concerning the reliability of applying a strontium isotope analysis to a forensic investigation. The regional geographic information gained from strontium isotopes can potentially be used to assist in identifying an individual through his/her region of origin.

Positive identification of skeletonized human remains is a difficult task when dental records and/or DNA are unavailable. Through archaeological research, strontium isotope analysis has successfully been used to trace individuals back to their place of birth using cortical bone and tooth enamel.¹ In forensic anthropological contexts, this method has the potential to help narrow down the search for missing persons to their geographical region of origin.² While strontium isotope analysis may be of use to forensic professionals, few studies have examined regionally-specific strontium isotope ratios among modern Americans.

This study utilized dental enamel from teeth of 76 individuals living in the New England region of the United States. Thirty-five of the samples were obtained from individuals donated to the Anatomical Gifts program in the Department of Anatomy and Neurobiology at Boston University School of Medicine. Forty-one samples were collected through individual donations from pre-arranged standard-of-care surgical dental procedures approved by an Institutional Review Board (IRB). In addition to birthplace, the type of water consumed during childhood was documented for the collected samples. The birthplaces represented by these individuals include the greater Northeast and Midwest of the United States (n=53), Central America (n=6), Caribbean Islands (n=7), West Africa (n=5), and Europe (n=3). Local faunal and water samples were collected from the New England region for local range comparisons. The samples were cleaned and approximately 10mg of enamel was removed from each tooth, acid washed, dried, and dissolved in nitric acid before analyzing the samples using a Thermal Ionization Mass Spectrometer (TIMS) for analysis of ⁸⁷Sr/⁸⁶Sr ratios.

The human ⁸⁷Sr/⁸⁶Sr ratios ranged from 0.705736 to

0.711876 and were grouped by geographical region. An analysis of variance was used to test for regional variation and significant differences were found. The samples from the United States were significantly different from the samples in Central America, Caribbean Islands, West Africa, and Europe. Central American samples were also significantly different from the other groups. No significant differences were observed between the Caribbean Islands, West Africa, and Europe. A significant difference was observed between bottled and tap water consumed by individuals from West Africa. The New England faunal samples from Pembroke, MA, and water sample from Braintree, MA, were not significantly different from the New England human samples, but the Brighton, MA, water sample was significantly different.

Based on the acquired data, regional differences in ⁸⁷Sr/⁸⁶Sr ratios are detectable using strontium isotope analysis. The results suggest that the differences observed are due to a combination of geological effects and influences from the globalization of food. The three distinct strontium ranges found across the five geographical regions correlated with the differing surface geology, yet slight differences in geology between the Caribbean Islands, West Africa, and Europe were not detected by strontium ratios. This suggests that region of origin may not be as reliable when analyzing only strontium isotopes in tooth enamel.

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Teeth, Isotopes, Human Identification

H64 Investigation of the Potential for Use of 3D Topographical Data and Geographical Information Systems for Age-at-Death Determination From Pelvic Skeletal Remains

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After attending this presentation, attendees will have been introduced to the use of Geographical Information Systems (GIS) and the approach of considering skeletal remains as "geographical landscapes" to explore methods of age-at-death determination. Attendees will have seen that the results suggest promising potential for further study and will understand some of the issues faced by the approach, such as standardization of orientation and mitigation against variations in size.

This presentation will impact the forensic science community by: (1) serving as a springboard for further research; (2) contributing to the understanding of age degeneration of pubic symphyseal and auricular surface (pelvic) features; and, (3) encouraging future development of age-at-death determination methods.

The use of the pubic symphyseal and the auricular surfaces

in methods of determining age-at-death from skeletal remains are well established, standard, and are commonly used. Current methods involve recording the phase that best corresponds to the specimen being analyzed, using a set of reference standard phase images and descriptions and visual observation of several features. While they have been shown to have a good level of accuracy, such methods inevitably involve an element of subjectivity and results are somewhat dependent on observer expertise. This study explores the potential of a quantitative method of determining age-at-death using parameters obtained from analysis of topographical data within a Geographical Information System (GIS). Pubic symphyseal and auricular surfaces from skeletal remains from a known sex and age-at-death collection (St. Bride's Church Crypt Assemblage, London, UK) were scanned to produce x, y, z point cloud data. The study sample included scan data from the os coxae of 89 individuals (49 left-side and 40 right-side), an age range of 17 to 91 years of age-at-death and the male to female ratio was 1.5:1 (54 males, 35 females). The 3D data from the regions of the pubic symphyseal and auricular surfaces were explored as "geographical landscapes" within arcGIS (ESRI® software). For the pubic symphysis surface, the relationship between age-at-death and the parameters (mean slope and mean aspect) were investigated. In general, values of mean slope and mean aspect, across the pubic symphysis surface, decreased with increasing age-at-death. These correlations were shown to be significant for the male study sample ($p < 0.05$). However, a similar correlation for females was not observed, possibly due to the smaller surface size and confounding contributions to pubic symphyseal surface features inherent to the female biological function of the pelvis. For the auricular surface, the relationship between age-at-death and the parameters (mean elevation, mean point density, and mean slope) were investigated. In general, values of mean elevation decreased with increasing age-at-death while, values of mean point density and mean slope increased with age-at-death. The correlation between mean point density and age-at-death was shown to be statistically significant ($p < 0.05$) for the whole study sample (sexes combined). The work presented represents a preliminary study but clearly indicates that there is the potential for the development of such work to generate a reliable, quantitative method for determining age-at-death from skeletal pelvic remains using GIS. Further work is recommended and this should include increasing the sample size and, in particular, the number of female specimens.

Age-at-Death, Pelvis, GIS

H65 Strontium Isotopes and Geolocation: The Pathway for Identification of Victims in Medellín, Colombia

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After attending this presentation, attendees will understand the necessity for new ways of identification of victims in Colombia, have a better understanding of strontium isotope analysis, and understand that isotope analysis on modern populations has limitations and that more research is needed.

This presentation will impact the forensic science community by serving as a starting place for future research with strontium isotopes on modern populations.

The goal of this study was to determine if strontium isotope analysis ($^{87}\text{Sr}/^{86}\text{Sr}$) is useful when applied to modern dental enamel

samples, specifically from Medellín, Colombia. Strontium isotope analysis has been used extensively in the archaeological literature with only a few studies in the forensic literature. A preliminary study by Juarez was one of the first to use modern samples from Mexico to determine the origin of border crossers.¹ As with that, there is a need for identification of individuals whose remains are found in Colombia, due to the conflict that has taken thousands of lives. Samples for this study consisted of 75 teeth from 61 individuals drawn from the Antioquia Modern Skeletal Reference Collection. This collection was developed to advance the knowledge of the Colombian population by developing standards and validation studies. The dental enamel samples were prepared using the procedures laid out by Balasse *et al.* to remove organics and adsorbed carbonates.² The samples were then transferred to a Thermal Ionization Mass Spectrometer (TIMS) at Boston University in the Earth Science Department, in order to run strontium columns to collect any strontium in the sample and then to run the sample through the TIMS. Standards and blank samples were run along with the study samples to test cleanliness of the equipment. The overall mean ($^{87}\text{Sr}/^{86}\text{Sr}$) for all samples in the study are 0.70739 +/- 0.00159. The samples were grouped by department, but due to the small sample size of many of the departments, the samples were grouped into the department Antioquia and all other samples. The overall strontium ($^{87}\text{Sr}/^{86}\text{Sr}$) mean of Antioquia samples is 0.70746 +/- 0.00174, and Non-Antioquia sample mean = 0.70704 +/- 0.00104. A Mann-Whitney U test, which is the non-parametric equivalent of a *t*-test, determined there is no significant difference between these two groups. The data was divided into samples from the city of Medellín against samples from other cities, or Non-Medellín; these groups have strontium isotope ($^{87}\text{Sr}/^{86}\text{Sr}$) means of 0.70748 +/- 0.00207 and 0.70710 +/- 0.00105, respectively. It was determined that these groups are not significantly different. Overall, the results are inconclusive due to small sample sizes. More samples from other geographic locations in Colombia are needed to accurately sort populations in Colombia. There are numerous factors that affect the strontium isotope signature in teeth, and modernization, specifically imported food and bottled water, has an effect on strontium isotope ratios in dental enamel of individuals. In order for this method to be either disregarded in forensic studies or utilized in forensics, more research with other modern populations needs to be performed.

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Strontium, Colombia, Isotopes

H66 Elemental Analysis of Cremains Using X-Ray Fluorescence Spectrometry

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After attending this presentation, attendees will understand the results of a study conducted to determine whether X-Ray Fluorescence (XRF) spectrometry may be a useful tool in the analysis of potentially contaminated cremains.

This presentation will impact the forensic science community by providing information regarding a new analytical

approach that could be used in the examination of cremated materials, especially if they are suspected of being contaminated with nonskeletal material.

Cremation can be achieved in various contexts including funerary practices as well as accidental or criminal incineration. During any process of cremation, contamination of the cremated remains is possible; this can happen naturally or unintentionally through taphonomic processes, or it can occur because of intentional improper cremation practices. Due to a number of criminal cases in which cremated remains have been contaminated or completely replaced with non-skeletal material (including the highly publicized Tri-State Crematorium in Nobel, Georgia involving the improper cremation of more than 300 bodies), it has come to the attention of investigators and forensic anthropologists that there is a lack of analytical methodology available for the examination of potentially contaminated human cremated remains. Especially with the growing efficiency of pulverizing machines used to grind cremated bones down to minute fragments and a fine powder, there is a need for instrumentation that can provide accurate analysis of these cremated remains to test for the presence of non-skeletal contaminants.

This study tested the ability of XRF to detect the presence of contaminants in cremated remains by assessing the elemental constituents of the sample. Specifically, a Bruker Tracer III-SD handheld XRF was utilized. The handheld XRF system has been developed as a general tool for crime scene investigation and, because of its portability, can be used to assess the elemental composition of evidence at the scene. Moreover, there is relatively little training required to use the instrument (although significantly more training is often required to properly interpret the results). A laboratory experiment was designed involving 11 samples of cremated remains of a standard poodle which were variably contaminated with concrete mix brand/type not listed or disclosed and then analyzed using the XRF. Bone is primarily comprised of the elements calcium and phosphorus. Calcium is also a significant component of typical concrete mixes, but it also contains other elements such as iron, silicon, and aluminum. Samples 1 and 11 contained 100% cremated remains and 100% concrete mix, respectively, and samples 2-10 successively decreased in cremated remains and increased in contaminant by 10%.

Results show that the elements detected in the samples were consistent with elements that comprise skeletal material and the contaminant used, and that the varying amounts of these materials in the test samples could be detected in the elemental photon counts of the instrument. The most significant changes were noted in the elements phosphorus, potassium, aluminum, iron, sulfur, and silicon. As the amount of cremated remains decreased, so did the levels of phosphorus and potassium. With increasing concrete mix, the levels of aluminum, iron, sulfur, and silicon increased.

It is therefore concluded that the handheld XRF is able to detect changes in the variable presence of cremated remains and the contaminant of the study samples. This approach can assist the forensic community by aiding forensic anthropologists and other analysts when examining cremated remains for the presence or degree of non-skeletal contaminant as well as assisting investigators by indicating possible improper or criminal cremation practices.

Cremains, Cremation, X-Ray Fluorescent Spectrometry

H67 Application of X-Ray Fluorescence for Sorting Commingled Human Remains

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After attending this presentation, attendees will gain an understanding of the application and reliability of portable X-Ray Fluorescence Spectrometry (pXRF) for differentiating skeletal elements of different individuals.

This presentation will impact the forensic science community by highlighting the application of pXRF in forensic anthropology. The challenges and limitations of this method for resolving cases involving commingled remains will be discussed.

Commingling of human remains presents complex challenges for forensic anthropologists. In such cases, it is important to accurately segregate each individual represented and determine the number of individuals present. For small-scale commingling cases, pXRF spectrometry provides a rapid and non-destructive method for potentially determining whether a set of remains belongs to a single or multiple individuals by analyzing trace element concentrations in human bone.

The goal of this study is to determine if pXRF can be used to reliably sort commingled remains. X-ray Fluorescence is capable of detecting minute differences in elemental concentrations, which is essential for discriminating between bones of different individuals. In order for the method to be useful for resolving commingling, there must be more variation in trace element concentrations *between* individuals than *within* individuals. To test this hypothesis, pXRF analysis was conducted on ten bones and one tooth from 20 human skeletons curated at the California State University, Chico's Human Identification Laboratory. From each individual, trace element concentrations were measured on the maxillary first molar enamel, the mental eminence of the mandible, the shaft of the fourth rib, the vertebral body of the fifth lumbar, and the midshaft of the humerus, ulna, radius, femur, tibia, and fibula. These elements represent the major anatomical regions of the skeleton and should account for expected variation within a single individual. Each sample was scanned without a filter for 180 seconds, and elemental concentrations were measured in reference to a mudrock calibration file. Slight differences were detected in comparisons of data from scans of cleaned versus uncleaned cortical bone surfaces. To reduce possible surface contamination effects, all bone surfaces were cleaned with 100% ethanol (EtOH) prior to analysis.

Elements detected included sodium, magnesium, aluminum, silicon, phosphorous, sulfur, potassium, calcium, barium, titanium, vanadium, chromium, manganese, iron, cobalt, nickel, copper, and zinc. The elemental concentrations from each bone and tooth were analyzed using discriminant function analysis. One of the ten skeletal elements from each individual was assigned group membership (cross validated). Finally, the potential influence of diagenesis was examined by comparing variation within and between individuals who were processed as relatively intact bodies versus those that were either recovered as skeletal remains from outdoor surface environments or from buried contexts. Preliminary results indicate that there is more variation between skeletons than within skeletons, suggesting that pXRF has utility in resolving cases of commingling. Variation between skeletal elements from the same individual may reflect differences in remodeling rates between bones of varying density. The degree of overlap in trace elemental concentrations between some individuals suggests that the method has the greatest utility for small-scale commingling cases.

Forensic Anthropology, Portable X-Ray Fluorescence, Commingling

H68 Chemical Differentiation of Osseous Samples From Non-Osseous Materials Using Scanning Electron Microscopy-Energy Dispersive X-Ray Spectrometry (SEM/EDX) and Multi-Step Statistical Analysis

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After attending this presentation, attendees will have a better understanding of the benefits of utilizing Scanning Electron Microscopy-Energy Dispersive X-Ray Spectrometry (SEM/EDX) and multi-step statistical analysis to distinguish fragmentary osseous samples from non-osseous materials of similar appearance and chemical composition.

This presentation will impact the forensic science community by demonstrating that, when combined with a multi-step statistical analysis process, SEM/EDX is a practical method for distinguishing between fragmentary osseous samples and non-osseous materials of similar appearance and chemical composition in a forensic laboratory setting. This method involves simple sample collection and analysis and utilizes instrumentation currently possessed by many forensic laboratories.

While identification of osseous materials is generally established on gross anatomical qualities, highly fragmented or taphonomically altered materials are often problematic and alternative methods, such as histological or chemical analysis, must be utilized. Recently, chemical methods have been proposed to sort unknown materials according to their Ca/P ratios. Ubelaker et al. proposed using SEM/EDX to achieve this distinction and Christensen et al. have validated XRF spectrometry for this application.^{1,2} An alternative method of analysis involves performing Principal Component Analysis (PCA) on element spectra to classify unknown materials based on their trace element composition. Zimmerman et al. proposed the validity of this method with data obtained using XRF.³ Subsequently, performing PCA on elemental data obtained using SEM/EDX demonstrates potential for sorting capabilities.

Samples were prepared by extracting a small amount of material from each location of analysis using a Dremel® tool. The resulting powder was adhered to the SEM stub using a carbon dot, and a small piece of copper tape was placed on each stub to use in calibration. Twenty-six samples were analyzed, with five spectra taken at each site, resulting in a total of 130 spectra. Samples consisted of human and non-human osseous materials (bone, antler, ivory, and shell), non-biological specimens (rock apatite and synthetic hydroxyapatite), and other biological specimens (sand dollar, shell, and two coral species). Data was collected using a LEO 1450VP Scanning Electron Microscope and an Oxford® Energy Dispersive Spectrometer. After all five spectra were collected for a sample, the detected elements were assessed. Carbon was removed due to suspected contamination from the stub as well as elements appearing in two or less spectra per sample. The remaining elements were normalized. Weight percent data was then processed in R, version 3.0.1, by the R Foundation for Statistical Computing using PCA, Linear Discriminant Analysis (LDA), and Quadratic Discriminant Analysis (QDA) based on principal components representing 99% of the variation within the data.

A three-tiered analysis was undertaken to improve

discrimination between sample groups. The first tier involved distinguishing between marine and non-marine samples. This resulted in 100% classification using LDA and QDA and was successful in discriminating octocoral, which has proven difficult when using Ca/P ratio-based analysis.^{1,2} Tier two analyzed only the non-marine samples indicated in tier one, dividing them into osseous and non-osseous materials. Classification was 100% using LDA and 97% using QDA with three rock apatite spectra classifying as osseous. Finally, tier three assessed osseous materials to determine if human and non-human samples could be distinguished. Results indicated 58% average classification between human and non-human osseous materials using LDA, with 80% of human samples misclassifying as non-human and 19% of non-human samples misclassifying as human, and 70% average classification using QDA, with 40% of human samples and 25% of non-human samples misclassifying. Misclassification may be a result of the large variety of species with limited representative samples and discrimination is anticipated to improve with a larger data set. Additionally, alternative statistical methods are being considered to improve classification.

Multi-tiered statistical analysis of elemental composition acquired using SEM/EDX demonstrate the ability to discriminate between osseous and non-osseous materials and show potential for differentiating between human and non-human samples. Further research will expand the data set by adding dental materials and additional samples chemically similar to bone in an effort to increase the accuracy of human and non-human differentiation and create a standard reference set to allow for use of this method in the forensic laboratory setting.

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Forensic Science, Elemental Analysis of Bone, SEM/EDX

H69 A New Approach to Radiographic Comparison: Combining Morphometric and Morphoscopic Techniques

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After attending this presentation, attendees will gain a better understanding of how morphometric and morphoscopic techniques can be combined to enhance the efficiency of visual radiographic comparison methods for human identification. The goals of this presentation are to: (1) evaluate the reproducibility of a morphometric comparison method that can be utilized to abbreviate a lengthy list of potential matches; and, (2) apply the morphometric method in constructing a test to evaluate the morphoscopic comparison method.

This presentation will impact the forensic science community by promoting a new approach to radiographic comparison that will enhance the efficiency of identifying unknown individuals when the size of the potential-match list precludes a manual search.

Prior studies have validated the use of chest radiograph comparison as a method of identification, and recent morphometric approaches have used Elliptical Fourier Analysis (EFA) to examine whether clavicular morphology is diagnostic for individuation. A new metric method based on clavicular shape outline has recently been developed and validated for use at the Joint POW/MIA Accounting Command, Central Identification Laboratory (JPAC-CIL) to expedite identifications made through radiograph comparison.¹ The present study serves to strengthen the protocol of this new method.

This method uses EFA to compare clavicle shape from 3D surface scans of Postmortem (PM) human remains with 2D hand-traced outlines from Antemortem (AM) radiographs by ranking individuals using shape similarity. Because this requires manual tracing of 2D clavicle outlines, its accuracy relies upon outline reproducibility. A series of observer-variation studies were conducted to examine potential measurement uncertainty associated with tracing of clavicle outlines and trimming of the medial and lateral ends. Repeatability was assessed using Coefficient of the Variance of the Error (CVE) and by comparing the EFA ranks generated for each outline. CVE results indicate that measurement uncertainty is relatively low (<15%). EFA results demonstrate small measurement differences between observers, with the correct match ranking in the top 5% of the sample 72% of the time, and in the top 25% of the sample 94% of the time. These results are consistent with a recent study.¹

The morphometric method was used to construct a morphoscopic test that presented examiners with a series of arrays containing images that varied in degree of morphological similarity, and thus difficulty. Degree of similarity between AM/PM images was defined through specific EFA values. AM radiographs were then morphoscopically assessed to ensure similarity in vertebral morphology. Radiographs were acquired from the medical records of unaccounted for U.S. service personnel located at the JPAC-CIL. The test consisted of five arrays, each containing one PM and five AM radiographs.

The test was administered to 42 individuals at the JPAC-CIL. Participants were asked to compare each PM image to the five AM images to determine whether a match existed. Overall rates are as follows: accuracy 85%; positive predictive value 73%; negative predictive value 94%; sensitivity 82%; and specificity 87%. The results were broken down based upon level of education, professional experience, and prior radiographic experience. In general, method performance tended to improve as level of education increased. However, participants with an MA/MS performed slightly better than those with a PhD. Participants with 10+ years of professional experience performed at higher rates than those with less experience. Interestingly, participants with no professional experience performed quite well as a group, generating higher rates than individuals with nine or fewer years of experience. Lastly, participants with prior training/experience in radiograph comparison outperformed participants with no prior training/experience.

Test results demonstrate the value of the morphoscopic comparison method for identifying unknown individuals, especially when employed by experienced analysts. Observer-variation studies of the morphometric method indicate method reliability, and when used in conjunction with the morphoscopic method, can enhance the efficiency of identifications by shortening the potential-match list. While results demonstrate the value of combining morphometric and morphoscopic methods for identification of individuals using chest radiographs, further research could investigate the applicability of this technique using skeletal elements from other body regions.

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Chest Radiograph, Elliptical Fourier Analysis, Positive Identification

H70 Forensic Sciences and the Duty of Memory: Identification of Two Civilian Heroes From the Second World War

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The goal of this presentation is to provide attendees with an example of successful interdisciplinary collaboration to clarify the fate of a group of missing civilians involved in French resistance movements during World War II.

This presentation will impact the forensic science community by highlighting the strategy and methodologies developed to trigger identification of incomplete, degraded skeletal remains and the contribution of forensic science to bringing closure to the victims' families, who have been waiting for 70 years.

On July 27, 1944, a group of civilians linked to French resistance movements was executed by Nazi soldiers and buried in a small wood near Toulouse. Despite being severely injured, one of them managed to escape and informed the families. Three months later, once the Nazis departed, relatives recovered 15 bodies; 10 were identified by their families. The five remaining bodies were first buried in individual graves, but in 1990, the bodies were re-exhumed and re-buried in a single mass grave. Based on archival investigation, a list of possible identities was presented by historians and family relatives to a prosecutor who requested an exhumation.

In 2011, the mass grave was exhumed and human remains were transferred to a field forensic facility to be examined by a pathologist and an anthropologist. The first steps were to establish the minimum number of individuals (MNI=5) and to isolate the best samples for DNA analyses. Because of the incompleteness, fragmentary nature, and poor preservation of skeletal remains (the grave was regularly flooded), teeth were collected from two maxillae and three mandibles and the best preserved long bones (femur and tibia) were also sampled to increase the number of possible profiles to be extracted.

The results of anthropological examination were consistent with historical investigation by determining the presence of five males with ages ranging between 20 and 45 years who had sustained a number of peri-mortem ballistic injuries and showed evidence of blunt trauma with recent bone remodeling. Personal belongings found associated with the skeletal remains were also consistent with information given by relatives and cellmates.

The DNA extractions from the teeth and long bones were performed using protocols developed to enhance the retrieval of degraded DNA molecules from ancient remains. The analytical strategy adopted was then to replicate the analyses and to combine the use of multiple commercial multiplex kits (AmpFISTR® Identifier® Plus™, AmpFISTR® Minifiler™, Investigator® IDplex™, PowerPlex® 21 and PowerPlex® Y23) to obtain the most complete DNA profiles. Two full and two partial DNA profiles were obtained from the 15 skeletal elements sampled and were compared to those of the relatives. To improve the discrimination power of the analysis, this study also used a commercial kit containing twenty-three Y-STRs (Y-23) to establish Y-haplotypes and compare the

male lineages. A partial Y haplotype was obtained and compared to the Y haplotype of the putative son.

Two of the men were positively identified via DNA. One was a Belgium former aviator who resigned in 1940 from his military position to join the Resistance where he created and led the Pyrenees Network. The second individual was leading a group of members of the resistance in the Toulouse region. The multidisciplinary work performed by historians, forensic anthropologists, pathologists, and biologists not only allows families to recover remains of their relatives but also to open the possibility for a national judicial investigation on war crimes. To date, historians are looking for the victim's descendants to be compared with the remaining profiles. More field research is ongoing in the same location to recover another group of missing civilians executed under the same conditions but without any witnesses.

Identification, DNA, Second World War

H71 Application of Stable Isotope Forensics for Provenancing Human Remains Recovered From Southeast Asia and the Pacific

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The goal of this presentation is to highlight the value of stable isotope forensics for provenancing human skeletal remains from unknown contexts. After attending this presentation, attendees will gain an understanding of the potential value of stable isotope forensics and how these approaches complement other human identification methods.

This presentation will impact the forensic science community by highlighting the potential of stable isotope forensics in provenancing human remains to aid in forensic identification.

The application of stable isotope analysis has provided novel approaches for provenancing unidentified human remains from forensic contexts. Stable isotope values measured in human tissues (e.g., bone, teeth, hair, and nails) provide a record of an individual's life history and location. Human bone stable carbon and nitrogen isotopes of collagen and stable carbon isotopes of bioapatite reflect consumption of food resources, which vary between geographic regions due to cultural dietary differences. Stable oxygen isotopes of bioapatite vary between regions due to environmental factors (e.g., aridity, elevation, and distance from large bodies of water) that influence the isotopic composition of drinking water.

This study discusses the application of stable isotope forensics for provenancing human bone obtained by the Joint POW/MIA Accounting Command, Central Identification Laboratory. Stable isotope analysis of human bone was used to determine whether U.S. Americans could be distinguished from Southeast Asians. More specifically, isotope values consistent with consumption of a strictly C₃-based diet (e.g., rice) were considered to be more likely associated with individuals from Southeast Asia, whereas values consistent with greater consumption of a mixed C₃/C₄ diet (e.g., corn, sugar) were considered to be more likely from North America.

The study included 30 human bone samples recovered from various sites in Southeast Asia and the Pacific, which were selected from known incidents and had a known number of decedents (i.e., while the exact identity of each individual may not be known, the origin of the individual is known — either U.S. American

or Southeast Asian). mtDNA and haplogroup assignments by the Armed Forces DNA Identification Laboratory were available for 27 of the samples; for three additional samples, other information pointed to the origin of the individual. The database included 27 samples and the remaining three samples were used as a hold-out test case.

Stable carbon isotope values of bone collagen varied from -20.5 to -14.7‰ (mean -17.4‰ ± 2.1), and stable nitrogen isotope values varied from 9.2‰ to 14.2‰ (mean = 11.9‰ ± 1.1). Bioapatite values varied from -17.1 to -8.9‰ (mean = -13.2‰ ± 2.6) for stable carbon isotopes and -9.9 to -4.9‰ (mean = -7.4‰ ± 1.0) for stable oxygen isotopes. The stable carbon isotope values of both collagen and bioapatite form a bimodal distribution and indicate diets consisting of varying amounts of C₃ and C₄ resources. When compared by known provenance, there was minimal overlap in collagen and bioapatite stable carbon isotope values between U.S. Americans and Southeast Asians. As expected, U.S. American stable carbon isotope values were significantly elevated relative to Southeast Asians, reflecting greater contribution of C₄ resources in the diet. Linear discriminant function analysis correctly classified 96.3% (cross validated) of the samples based on stable carbon isotope values. Stable nitrogen isotope values of bone collagen show much lower variation and were not significantly different between groups.

Three samples, representing different skeletal elements from the same individual, were selected as a test case. Stable carbon isotope values were nearly identical; importing the sample and study data into FORDISC® 3.0, custom discriminant functions were generated for analysis. Stable carbon isotopes of collagen classified the samples as U.S. American with posterior probabilities between 0.711 and 0.557. Carbon isotopes of bioapatite yielded similar results. Cross-validated accuracy rates were 96.3% with this model. The addition of stable nitrogen isotopes to the analysis still classified the samples as U.S. American, but with lower posterior probabilities.

Looking forward, isoscape models will next be used to predict possible areas of geographic origin within a specific region based on the spatial distribution of oxygen isotopes in drinking water. This study has demonstrated that stable isotope analysis provides valuable information for estimating the provenance of human remains and can aid in eliminating samples from consideration that are unlikely to be of forensic significance.

Forensic Anthropology, Stable Isotope Analysis, Provenancing Human Remains

H72 The Exhumation and Interdisciplinary Scientific Analysis of a World War II Mass Grave From Hungary

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After attending this presentation, attendees will better understand the importance and value of interdisciplinary scientific analysis in the excavation and methods of identification of individuals recovered from war graves.

This presentation will impact the forensic science community by demonstrating how multiple disciplines can be brought together in order to achieve a common objective — the scientific analysis of a World War II mass grave. The advantages of combining archaeological research and exhumation, anthropological analysis, museological studies, and military history will be presented through a case study that illustrates how each

discipline can contribute.

During WWII, tens of thousands of soldiers died in Hungary. After the autumn of 1944, troops rarely had time to document their losses and many individuals were buried in unknown locations. Current soil exploratory work, farming, and construction work often expose World War II artifacts, explosives, and human remains on the ground surface. This is exactly what occurred at the end of May 2011 in Pettend, Fejer county, where a mass grave was unearthed. The primary objectives in the analysis of the material and skeletal remains recovered from this mass grave were to identify the nationality of the remains if possible, and use anthropological methods to separate commingled remains and determine if any identifying characteristics (antemortem injuries, etc.) were present, that may be unique to a particular individual and potentially useful for future individual identification. These objectives were met through the interdisciplinary nature of the analysis.

During the exhumation of the grave, an odd positioning of the remains was observed. At the lower layer of the grave rested the remains of three soldiers with their bodies placed in a parallel plane. Directly on top of them, three individuals had been buried in two layers in a more uncharacteristic way (one of them was laid facedown). The excavation also uncovered a number of isolated commingled human remains. The anthropological analysis of these bones identified at least three additional individuals besides the initial six. This burial manner was informative as to who might have buried the dead and, in turn, who the dead might be. The haphazard placement of the individuals in the grave refers to an iniquitous funeral, which suggests that either there was not enough time to carry out a proper funeral or respect was deliberately not given to the deceased. Knowing what we know about wartime behavior and historical incidents of burial, especially in mass graves, this case appears to be indicative of a burial carried out by enemy combatants. The items of clothing (boots, buttons) and the type of ammunition and grenades found with the remains were consistent with those used by the contemporary Soviet Red Army. Researching the military history of the area during World War II revealed that Soviet soldiers had been fighting in the region December 7-22, 1944 and January 24-28, 1945. The soldiers lying in the grave in Pettend could have fallen during any of those days. The temperature was below freezing throughout both of these months, so there is a good chance that the burial of these men happened long after their deaths.

Aside from the information gleaned from the grave itself, the skeletal and cultural material provided evidence concerning trauma. Traces of injury were isolated during the exhumation in the form of in situ shrapnel found with the remains, and in one case a Hungarian machine gun bullet tip was observed between two vertebrae. Anthropological analysis also found peri-mortem trauma that was consistent with the type and position of the aforementioned recovered firearm remnants. Finally, one case revealed evidence of antemortem trauma that might be useful for future individual identification.

The interdisciplinary analysis of the World War II mass grave excavated in Pettend provided a number of scientific results. Although the individual identification of the skeletal remains has not yet occurred, the number of individuals interred, the circumstances surrounding their deaths, the relatively precise time period when they died, as well as their service of duty was revealed. As a result of the conclusions drawn from the analysis, the Hungarian Department of Defense, Division of Public Relations and War Grave Office was able to repatriate the remains of the soldiers to the national military cemetery in the presence of adequate representation.

Interdisciplinary Scientific A, World War II, Mass Grave

H73 Stature Estimation in the CIL: Is There Significant Noise in the Trotter and Gleser Data?

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The goal of this presentation is to educate the attendees on special issues concerning the most important stature estimation formula in forensic anthropology.

This presentation will impact the forensic science community by improving awareness of problems that will affect stature estimation with the popular Trotter and Gleser models.

Stature estimation is one of the basic components of the biological profile of skeletal remains. The stature estimate is used as part of a general description of the physical characteristics of the deceased or, when the antemortem stature of a missing person is available, it can be formally compared by means of a statistical test. Either approach contributes to the goal of identification of the deceased.

Stature estimation has arguably seen the most aggressive utilization in the various Central Identification Laboratories (CIL) operated by the United States military since the Second World War. It was in the immediate post-war era that the Army began to utilize professional anthropologists for the purposes of identifying skeletonized remains of U.S. service personnel who died in combat but whose bodies were not recovered until months or even years later. Since actual measured statures had been recorded at induction for every missing soldier, the anthropologist who developed the identification procedures made systematic use of stature when comparing skeletal characteristics to the profiles of the missing. It was in this context in 1948 that Mildred Trotter recognized the need for new models based on reference data from the very population to which the models were to be applied. This data was to come largely from the CIL cases being identified in Hawaii, but augmented with data from the Terry Collection. Trotter collected data from the CIL cases, took measurements from the Terry Collection, and partnered with Goldine Gleser to calculate the regression models that are well-known to forensic anthropologists around the world.

The standard operating procedures in use in the Hawaii CIL in 1947-48 directed that measurements and observations were made by soldiers who were supervised by an anthropologist. The laboratory operation was large and there was only a single anthropologist employed. One consequence of this situation was that Trotter did not measure the military cases included in her reference data. She was responsible for ensuring that the soldiers knew how to take the measurements and she signed off on each skeletal analysis form that was completed.

Approximately ten years ago, anthropologists at the Joint POW/MIA Accounting Command (JPAC) CIL began to systematically examine old records from its precursor laboratory in the 1940s and from the Kokura Central Identification Unit (CIU) that operated in Japan during the Korean War. The opportunity to measure anew skeletal remains measured in the 1940s presented itself for several cases that had been disinterred for further testing. It became apparent that "standard" measurements of the femur and tibia were not being taken in a consistent manner. There appeared to be some differences among the anthropologists who signed the skeletal forms. Further, examination of the Standard Operating Procedures manuals from Hawaii (1948) and Kokura (ca. 1950-55) revealed that the intent was to take the physiological lengths of the femur and tibia for use with Rollet's (1888) stature tables (or formulae based on them) as opposed to the total lengths indicated for use in the Trotter and Gleser 1952 paper. The forms used in the

1940s simply labeled the measurements as “Femora” for the length of the femur and “Tibiae” for the length of the tibia.

Bivariate plots of data taken from forms signed by Charles Snow, Alexander Tardy, and Mildred Trotter were examined to determine which measurements were actually being recorded on the forms. Specifically, the raw data for the femur and tibia, respectively, were plotted against the fibula length since the length of the fibula had been measured in a single manner throughout the 20th century. This raw data was compared to a regression model line (with fibula as dependent variable) based on reference data measured in recent years in the CIL or from the Forensic Databank from the University of Tennessee, Knoxville. The data in the plots revealed that the femur and tibia measurements are too short relative to the fibula to have been consistently taken as total lengths (e.g. the residuals are biased). The spread of the raw data also *suggests* that the femur and tibia were measured inconsistently, as in sometimes total length and sometimes the physiological length. After the data had been plotted, the historian Heather Harris shared a letter from Goldine Gleser to Mildred Trotter dated April 7, 1957, in which Gleser suggests to Trotter that the femur had not been consistently measured as total length in the various laboratories that had contributed to their reference data. She alluded to a larger problem with the tibia. Of course, Gleser the statistician was noting a problematic pattern in the data.

What difference does noisy data make? This study calculated new regression models for the femur and tibia based on modern CIL data combined with the Forensic® Data Bank. Statures were estimated for three resolved CIL cases with the new models and compared with the stature estimates using the Trotter data in FORDISC® 3.0. Differences in the point estimate on the order of 0.5 inches can be expected when using the femur and on the order of 1.0 inch when using the tibia. These differences can have forensic significance. Consequently, it is proposed that the old CIL data be scrubbed and re-formed with modern CIL data so that only reliable measurement data are included. New models will be developed from the clean reference data.

Stature, Measurements, Trotter

H74 Building Standards and Capacity to Investigate Mass Graves in Iraq: Requirements and Challenges

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After attending this presentation, attendees will gain an understanding of the key challenges facing the forensic anthropologists and archaeologists of the International Commission for Missing Persons (ICMP) who are delivering training, introducing standard operating procedures, and building the capacity of Iraqi national teams to independently and effectively undertake mass grave excavations and examinations in post-Saddam Hussein-era Iraq.

This presentation will impact the forensic science community by demonstrating how effective assessment, planning, and flexible delivery based on legal and national requirements can achieve impact in the face of post-conflict, organizational, and logistical difficulties to successfully develop a national capacity to investigate and recover missing persons.

It has been widely publicized that up to one million Iraqis are currently missing within the country as a result of Hussein Regime crimes, mass executions, and cross-border wars. Many

thousands of these missing persons are believed to be buried in a vast number of mass graves which litter the cities, towns, and countryside. Since late 2008, the ICMP has worked to assist both the Iraqi government and, ultimately, the families of the missing by addressing the issue of mass graves in Iraq through an integrated program of training in forensic archaeology and anthropology, augmenting the Iraqi national capacity in mass grave excavation, and the examination of human remains.

The project objectives include assisting the Ministry of Human Rights (MHR), Medical Legal Institute (MLI), and Ministry of Martyrs and Anfal Affairs (MoMAA) in the implementation of investigative and scientific work that fulfils the requirements of the Iraqi Law on the Protection of Mass Graves and laws governing the roles of forensic doctors at exhumations. A further objective is improving the capacity of Iraqi professionals of all ethnic groups in locating, excavating, and examining mass graves and ensuring that the process is sustainable. The required outcome is that Iraqi teams develop an acceptable minimum standard of excavation, recording, examination, and evidence control that is implemented in investigations. This standard must be acceptable for courts and provide suitable data and samples for successful parallel DNA analysis, database, and antemortem data collection processes that can provide a basis for future identifications.

Assessment of existing capacity and institutional knowledge allowed planning of strategies to deliver a modular training program based on developed best practice and experience. A flexible system provided skills knowledge, demonstration and practice, assessment, feedback and post-course mentoring, examination, and reporting. The ICMP has employed a program of practical mentoring support to assist teams in gaining and sustainably maintaining the experience and skills to work independently to consistent required legal standards. Site investigation monitoring by ICMP provides dynamic progress feedback, allowing teams to develop and ICMP to design further training to enhance and focus on specific skills development. Reviews of ICMP training and monitoring of the post-course placement of trainees in their institutions provides data to allow flexible adaptation of courses to suit the requirements of ministries and teams. Further advanced training in new disciplines and skills (such as dealing with commingled remains) is being undertaken to ensure consistent standards are developed and maintained.

Maintaining momentum and meeting ambitious targets in a post-conflict environment poses many operational and strategic challenges for ICMP International training teams. This presentation offers an overview of the key challenges with particular focus on security concerns, procurement and logistics, environmental factors, and multi-agency coordination. The future requirements and the need to continue the development of their departments will be discussed. The scale of the potential project to identify the missing is a great challenge.

Mass Graves, Iraq, Standards

H75 Age Estimation in the Living: A Test of Six Radiographic Methods

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After attending this presentation, attendees will have an understanding of the different methods available for radiographic age estimation and the accuracy of these methods. This presentation will demonstrate the optimal method to use, given the proposed age of the individual.

This presentation will impact the forensic science community by enhancing understanding about the accuracy of methods of age estimation, which are already in existence, as well as proving to the court itself that radiographic methods of age estimation are reliable enough to be presented to a court of law.

Worldwide, over 51 million children are not registered at birth. The increase in cross-border movements in recent years, both for humanitarian and for economic reasons, has led to an increasing number of people who are either unable or unwilling to provide documentation providing proof of age. This has led to an increasing requirement for age estimation in the living for medicolegal purposes, especially among younger individuals since access to resources relies on their ability to prove age. Traditionally, age estimation in the living has relied on the use of left hand wrist radiographs which use skeletal maturational changes to estimate chronological age in the living.

This presentation will present research which tested radiographs of children of known age from a Scottish population against the commonly used atlases already in existence. The research examines the role of skeletal development in age estimation in relation to four topographical areas of the body: the hand/wrist; the elbow; the knee; and the foot/ankle. The accuracy and reliability of these age estimation methods and their importance in relation to the demands of modern-day practice will be discussed.

This research found that the use of knee and the corresponding atlas of Pyle and Hoer was as accurate for age estimation of females ($R^2=0.954$) and males ($R^2=0.952$) as the use of the traditional left hand-wrist radiographs and the Greulich and Pyle atlas (females $R^2=0.939$, males $R^2=0.940$).^{1,2} This underlines the ability of the practitioner to use radiographs which might already be in existence rather than having more radiographs taken. Finally, by comparing the results for all of the skeletal areas and corresponding atlases, it was possible to devise a guide showing which method is optimal for use in any given situation, allowing the practitioner to choose the most robust approach to age estimation possible.

References:

1. Pyle SI, Hoerr NL. A radiographic standard of reference for the growing knee. Springfield: Charles C. Thomas, 1969.
2. Greulich WW, Pyle SI. Radiographic atlas of skeletal development of the hand and wrist. Stanford: Stanford University Press, 1959.

Age Estimation, Radiographs, Children

H76 Comparison of the Use of Lamendin Variables and Aspartic Acid Racemization to Estimate the Age-at-Death

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After attending this presentation, attendees will gain a better understanding of the potential application of aspartic acid racemization to the estimation of age-at-death from teeth in comparison to the use of Lamendin variables.

This presentation will impact the forensic science community by providing the first comparison between the two techniques and illustrating their accuracy of age-at-death estimates in two Spanish populations.

One of the challenges in forensic investigation is to estimate the age-at-death of an individual. This is a fundamental parameter in mass disasters where the skeletons are often incomplete, which

makes the correct identification of the victims difficult. Teeth may be preserved long after all other tissues have disappeared and are often used to estimate characteristics like age-at-death.

There are several approaches to age estimation based on dental development. In forensic anthropology, the Lamendin technique and its variants are non-invasive methods for estimation of age-at-death. However, the method can only be used in single-rooted teeth. New methodologies for age estimation are based on the natural process of aging, which causes alterations of tissues and organs on different biochemical levels. The racemization of aspartic acid is one of these alterations. Although it is not exempt of disadvantages, such as the destruction of the sample teeth, its accuracy in the forensic age estimation has been demonstrated.

The aim of this research is to compare the use of Lamendin variables and aspartic acid racemization for the estimation of age-at-death and assess the reliability of these methodologies in two Spanish populations. Thirty-five healthy erupted third molars from Asturias, NW Spain, and thirty-five healthy erupted third molars from Cataluña, NE Spain, were collected from dental clinics. The Smithsonian Institution's ethical committee approved all procedures related to experimentation with human subjects. Root height, periodontosis height, and root transparency were measured three times in each molar using a digital sliding caliper. The average of these three measurements was evaluated in relation to the age. In both populations, periodontosis height, and root transparency showed a positive and significant correlation with the age, stronger in the Cataluña population. Since there is not a formula to estimate the age-at-death in multiradicular teeth, multivariate regression equations were generated for both populations based on these three parameters. Although both regressions produced highly significant F values ($p<0.01$), the r value was higher in the Cataluña population (0.86) than the Asturias population (0.78).

After taking these measurements, the teeth were cleaned and enamel and cementum were removed. The dentin was isolated, mechanically ground, and divided in aliquots of 200mg each. The dentin from five Cataluña third molars and from ten Asturias third molars was subjected to chemical derivatization to analyze the aspartic acid racemization ratios in Gas Chromatography/Mass Spectrometry (GC/MS). Unexpectedly, the racemization ratios were highly different between the two populations; the Asturias population showed double the racemization ratio than the Cataluña population in each age range. Despite these differences, a linear regression formula was generated to compare with the Lamendin Spanish regressions. Both populations had highly significant F values ($p<0.01$); the r value was higher than observed with the Lamendin variables, 0.996 in the Cataluña population and 0.959 in the Asturias population. The aspartic acid racemization showed a better correlation with the age than the approach using the Lamendin variables.

The accuracy of the two techniques in age estimation was investigated using statistical analysis. For the Lamendin variable methodology, the Medium Error (ME) in the Asturias population was -2.30, Standard Deviation (SD) 7.37, and Error Standard (ES) 2.60. In contrast, using aspartic acid racemization the following values were determined: ME, 0.062; SD, 4.80; and ES, 1.70. Again, the results were better in the Cataluña population, using Lamendin variable methodology: ME, 1.41; SD, 3.49; and, ES, 1.56. However, the best accuracy was found with the aspartic acid: ME, 0.0014; SD, 0.94; and, ES, 0.42. These results are in agreement with previous studies in which the ES using aspartic acid racemization was also low.

These research results provide a comparison between anthropological and biochemical methodologies. Although the aspartic acid racemization seems to be the more accurate technique, it is not exempt of drawbacks, and thus the anthropological technique represents a viable option.

Age-at-Death, Lamendin Variables, Aspartic Acid Racemization

H77 Temporal Characterization of Ossification of the Crania in Australian Subadults: New Standards for Age Estimation Using Computed Tomography

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After attending this presentation, attendees will gain awareness of the ontogeny of cranial maturation, specifically the fusion timings of primary ossification centers in the basicranium and the temporal pattern of closure of the anterior fontanelle, to develop new population-specific age standards for medicolegal death investigation of Australian subadults.

This presentation will impact the forensic science community by demonstrating the potential of a contemporary forensic subadult Computed Tomography (CT) database of cranial scans and population data to recalibrate existing standards for age estimation and quantify growth and development of Australian children. This research welcomes a study design applicable to all countries faced with a paucity in skeletal repositories.

Accurate assessment of age-at-death of skeletal remains represents a key element in forensic anthropology methodology. In Australian casework, age standards derived from American reference samples are applied in light of scarcity in documented Australian skeletal collections. Currently, practitioners rely on antiquated standards, such as the Scheuer and Black compilation for age estimation, despite implications of secular trends and population variation.¹ Skeletal maturation standards are population specific and should not be extrapolated from one population to another, while secular changes in skeletal dimensions and accelerated maturation underscore the importance of establishing modern standards to estimate age in modern subadults. Despite CT imaging becoming the gold standard for skeletal analysis in Australia, practitioners caution the application of forensic age standards derived from macroscopic inspection to a CT medium, suggesting a need for revised methodologies.

Multi-slice CT scans of subadult crania and cervical vertebrae 1 and 2 were acquired from 350 Australian individuals (males: n=193, females: n=157) aged birth to 12 years. The CT database, projected at 920 individuals upon completion (January 2014), comprises thin-slice Digital Imaging and Communications in Medicine (DICOM) data (resolution: 0.5/0.3mm) of patients scanned since 2010 at major Brisbane Childrens Hospitals. DICOM datasets were subject to manual segmentation, followed by the construction of multi-planar and volume-rendering cranial models, for subsequent scoring. The union of primary ossification centers of the occipital bone were scored as open, partially closed, or completely closed; while the fontanelles and vertebrae were scored in accordance with two stages. Transition analysis

was applied to elucidate age at transition between union states for each center and robust age parameters established using Bayesian statistics. In comparison to reported literature, closure of the fontanelles and contiguous sutures in Australian infants occur earlier than reported, with the anterior fontanelle transitioning from open to closed at 16.7±1.1 months. The metopic suture is closed prior to ten weeks post-partum and completely obliterated by six months of age, independent of sex. Utilizing reverse engineering capabilities, an alternate method for infant age estimation based on quantification of fontanelle area and non-linear regression with variance component modeling will be presented. Closure models indicate that the greatest rate of change in anterior fontanelle area occurs prior to five months of age.

This study complements the work of Scheuer and Black, providing more specific age intervals for union and temporal maturity of each primary ossification center of the occipital bone.¹ For example, dominant fusion of the sutura intra-occipitalis posterior occurs before nine months of age, followed by persistence of a hyaline cartilage tongue posterior to the foramen magnum until 2.5 years, with obliteration at 2.9±0.1 years. Recalibrated age parameters for the atlas and axis are presented, with the anterior arch of the atlas appearing at 2.9 months in females and 6.3 months in males, while dentoneural, dentocentral, and neurocentral junctions of the axis transitioned from non-union to union at 2.1±0.1 years in females and 3.7±0.1 years in males. These results are an exemplar of significant sexual dimorphism in maturation (p<0.05), with girls exhibiting union earlier than boys, justifying the need for segregated sex standards for age estimation.

Studies such as this are imperative for providing updated standards for Australian forensic and pediatric practice and provide an insight into skeletal development of this population. During this presentation, the utility of novel regression models for age estimation of infants will be discussed, with emphasis on three-dimensional modeling capabilities of complex structures such as fontanelles for the development of new age estimation methods.

Reference:

1. Scheuer L, Black SM. Developmental juvenile osteology. San Diego CA: Academic Press, 2000.

Subadult Age Estimation, Skeletal Maturation, Computed Tomography

H78 Age Estimation in the Indian Population by Ultrasonographic Evaluation of Apophyseal Ossification of the Iliac Crest

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After attending this presentation, attendees will learn to what extent Ultrasonography (USG) as a diagnostic modality will correlate with the present gold standard radiography for age estimation.

This presentation will impact the forensic science community by providing insight into the feasibility of ultrasound at the iliac crest for age estimation in continuation of earlier research studies and also the inherent benefits of Ultrasonography as an age estimation modality in the presence of statistically valid results.

Age estimation of the living has a long tradition in Medicine and is a very important task for a forensic practitioner in criminal and civil cases. Presently, radiography is taken to be

the gold standard for deducing age in the medicolegal age groups. However, it is associated with the inherent hazards of radiation. On the other hand, ultrasound is a radiation free, valid, rapid, cost-effective and non-ionizing imaging procedure which can be utilized for age estimation in the living while still relying on important skeletal maturity indicators concurrent with the chronological age. So, the present study was undertaken as an endeavor to compare radiographic evaluation of apophyseal ossification of the iliac crest with Ultrasonography for age estimation in the living.

The goal of this research is to study the sonographic evaluation of the apophyseal ossification of the iliac crest and to compare it with radiography in terms of sensitivity, specificity, and accuracy in the Indian population. Sixty subjects of both sexes, ranging in age from 10.6 years to 22.7 years, underwent Ultrasonography followed by X-rays of the iliac crest. X-rays were performed in the antero-posterior view followed by Ultrasonography scanning the entire length of the iliac crest. Age was deduced with the help of Ultrasonography and X-ray films by two separate, independent observers, in a double-blind manner. There was found an excellent degree of agreement and correlation between X-rays and Ultrasonography with a kappa value of 0.959 and p value of 0.995, respectively. Sensitivity and specificity of USG was 87.5 % and 94.1 % respectively. Accuracy was 92.0 %. Positive predictive value was 87.5%. Negative predictive value was 94.1%. It was possible to perform Ultrasonography for age estimation in all the subjects without any problems in general or particular. In view of the results obtained, it is concluded that Ultrasonography can be used to estimate age without undue risk of radiation. In addition to this, there may be a role for this new method as an adjunct to radiography for repeated measurements. By conducting more studies by Ultrasonography on a representative sample size, USG can be made a valid alternative to X-Rays for age estimation.

Ultrasonography, Iliac Crest, Age Estimation

H79 The Utility of Dental Crown Size in the Assessment of Ancestry

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After attending this presentation, attendees will understand how tooth crown size varies between sexes and broad geographically based groups.

This presentation will impact the forensic science community by identifying an additional method for the assessment of ancestry that can be adopted by the forensic anthropologist as part of the development of the biological profile.

Data were collected from the dentition of populations representing multiple geographic regions throughout the world (n>5,600). Buccolingual and mesiodistal crown measurements were taken for one side of the dental arcade, for a total of 32 metric variables per individual. Data were coded into one of three geographically based ancestral groups: African; Asian; and European. Data were then explored to identify differences between these groups. A general pattern was identified wherein African populations have the largest teeth, Asian populations are intermediate in tooth size, and European populations have the smallest teeth, although Asians tend to have slightly larger anterior teeth than Africans. Univariate Analysis of Variance analyses

(ANOVA) identified differences between the sexes in all metric variables except for three mesiodistal measurements on anterior teeth (upper second incisor, and lower first and second incisors). Observer error was evaluated in this study and found to be low, as reported in other studies using dental metrics.

A subset of this larger dataset, which includes only those individuals with complete dentition (n=508), was used to identify the efficacy of crown size in discriminating between ancestral groups. Models performed best when all teeth were included with the exception of the third molars, which were often missing or otherwise highly variable. Discriminant function analysis correctly classified individuals into one of the three ancestral groups, regardless of sex, in 71.3% of cross-validated cases. Further analyses identified sex differences between groups as the leading cause of misclassifications. Therefore, data were divided into ancestral group and sex. When sex is known, classification rates are improved; up to 88.1% of cross-validated cases in females, and 71.9% of cross-validated cases among males.

This study identifies broad patterns in crown size related to ancestry and sex. Dental measurements have the potential to be analyzed in a similar fashion to cranial measurements; therefore, a large database can be adopted for assessment of ancestry. Moreover, data on tooth dimensions can be quickly recorded with low observer error by individuals with only limited training. Based on these results, data on crown size can be included as part of the biological profile.

Dental Metrics, Ancestry, Discriminant Function

H80 Statistical Classification Methods for Estimating Ancestry Using Morphoscopic Traits

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After attending this presentation, attendees will understand the practical application of morphoscopic traits in ancestry assessment using a forensic dataset.

This presentation will impact the forensic science community by debunking some of the traditional approaches to ancestry assessment, which remain rooted in typological thinking. Moreover, this presentation will explore aspects of ancestry assessment method and theory.

Previous work has demonstrated the efficacy of morphoscopic (non-metric) traits in the assessment of ancestry, but only if the data are analyzed in a statistical framework and only if typological trait lists relying on extreme trait values are abandoned. There are numerous new statistical methods that can be used for the classification of an unknown cranium into a reference population on the basis of morphoscopic trait expression. Trait lists, however, remain the most widely taught, used, and accepted method of ancestry assessment. Trait lists are intuitively easy but lead to classifications without empirical support or consideration of the variation within groups. Without an explicit method, their validity is uncertain.

The objective of this study is to examine the utility of 16 frequently cited morphoscopic traits when analyzed using numerous statistical methods for classifying an unknown cranium into one of several reference groups based on those traits. The OSSA method and ten additional multivariate statistical classification methods (artificial neural networks, support vector machines, random forest, naïve Bayesian, decision tree, k-nearest neighbor, quadratic discriminant function, linear discriminant function, and

logistic regression) that provide multi-group classifications were tested using a large, modern sample of American Black (n = 256), American White (n = 218), and Hispanic (n = 244) individuals. The sample composition is particularly important to U.S. forensic anthropologists, as these groups represent the majority of the populations encountered during routine forensic investigations. Each of the statistical methods take correlations among variables into account as necessary and, more importantly, provides estimated accuracy rates for the classification of an unknown. All of these methods have different requirements for optimal classification accuracy of individuals; some methods require that the data have a multivariate normal distribution and others have virtually no requirements of the data. Rigorous cross-validation methods help to discern the best traits to use in classification and provide a measure of validity, that is, the expected accuracy of each classification method.

Artificial Neural Networks (aNNs), OSSA, support vector machines, and random forest models showed mean classification accuracies of at least 85%. The aNNs had the highest overall classification accuracy (87.8%) and random forests show the smallest difference between the highest (90.4%) and lowest (76.5%) classification accuracies.

The results of this research demonstrate that morphoscopic traits can be used to assess ancestry accurately, with low estimated error rates and without relying only on the experience of the observer, and that would be true if you showed someone else the traits and tested them. Moreover, these results demonstrate that subjective trait lists do not capture the true range of variation; each one of the classification methods has a firmer empirical basis for a classification than the traditional approach. There is a definite need for scientific rigor in ancestry assessment methods that are empirically supported by the data at hand. Every method, whether novel or long-established, should be tested and refined rather than performed simply due to tradition or subjective/personal experience.

Ancestry, Morphoscopic Traits, Classification Statistics

H81 Overjet or Prognathism? Morphometric Assessment of Midface Morphology Between Blacks, Whites, and Hispanics

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After attending this presentation, attendees will understand the utility of morphometrics to elucidate shape differences in the human palate and ultimately use this information as a resource to aid in ancestry estimation.

This presentation will impact the forensic science community by serving as a means to enhance the biological profile, specifically the estimation of ancestry, using a simple model that is easy to interpret.

In a recent paper by Hefner *et al.*, the ancestry of Black, White, and Hispanic crania were estimated through the combination of metric and non-metric observations through random forest models.¹ In their findings, Hefner *et al.* noticed a trend for the model to focus on maxillary, and specifically palate, variables as major discriminators.¹ This study noticed a trend for Hispanic crania to exhibit a pseudo-prognathism that was related to an orthodontic condition known as overjet, in which there is an increased projection of the teeth, labially or buccally, projecting at an angle anteriorly,

thus giving the appearance of prognathism. The aim of the current study is to refine the observations of Hefner *et al.* through the use of geometric morphometric analyses of recent Black, White, and Hispanic crania and to test which classification statistics are most appropriate for morphometric data, i.e., which methods yield the most optimistic classification results.¹

For the present study, coordinate data for 287 crania were collected. Coordinate landmarks were chosen to represent the midface adequately and included: alveolon; basion; bregma; ectomolare (left and right); prosthion; and, staurion. Data were obtained for American Whites (n=84) and American Blacks (n=82) from the Terry Collection at the Smithsonian and from the Pima County Office of the Medical Examiner in Tucson, AZ (n=121). The data were subjected to a Generalized Procrustes analysis that translated, scaled, and rotated the data. The raw coordinate data were transformed into Procrustes coordinates (size-free shape coordinates) and further refined using principal components analysis on the Procrustes coordinates to analyze the midfacial complex as an entire unit, in addition to simply x, y, or z coordinates.

Total correct classifications ranged from 65.5% to 89.3% depending on the statistical analysis used and whether Procrustes coordinates, or their derived principal component scores, were used. The single highest classification was achieved through random forest model using the Procrustes coordinates. In the random forest model using the Procrustes coordinates, Whites classified most accurately at 92.9%, followed by Hispanics at 90.1%, and lastly Blacks at 84.1%. Across the board, Blacks most commonly misclassified as Hispanic, while Hispanics generally misclassified equally as White or Black. Lastly, Whites tended to misclassify most commonly as Hispanic.

The analyses based on the Procrustes coordinates yielded higher overall classification results. It might be the case that generating principal components from Procrustes coordinates generates redundant data due to some anatomical structures being relatively stable between population groups. Ectomolare, basion, bregma, and prosthion overwhelmingly contributed to both the classification models, as well as generating the greatest loadings for each of the principal components. Principal component 3 contained 12.6% of the total shape variation and was consistently an important variable among all of the analyses. Principal component 3 demonstrated major shape changes in a reduction of palate width, with ectomolare moving anteromedially and prosthion moving distally. Additionally, staurion moved distally with the same magnitude as prosthion. Whites demonstrated the most exaggerated reduction in palate width relative to the cranium, while also demonstrating a deeper palate anterodistally. Blacks exhibited shape just the opposite of Whites with wider, shallower palates relative to the cranium, but with the addition of an exaggerated anteriorly projecting prosthion, related to the prognathism. Hispanics demonstrated an intermediate morphology with a slight reduction of palate width and depth, but with a slightly more exaggerated prosthion, likely the effects of overjet. The palate has proven to be an effective discriminator of ancestry, and with a simple model using morphometrics, fine-grained shape differences were observed.

Reference:

1. Hefner JT, Spradley MK, Anderson BE. Ancestry assessment using random forest models. *J Forensic Sci*, In press.

Forensic Anthropology, Geometric Morphometrics, Ancestry Estimation

H82 Exploring Chinese Cranial Affinity Within “Hispanic” Populations

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The goal of this presentation is to refine biological profiling for Hispanic populations using geometric morphometrics.

This presentation will impact the forensic science community by highlighting the importance of understanding biological variation in the determination of ancestry for populations broadly grouped as “Hispanic.”

Hispanics are the largest minority population in the United States, accounting for nearly 17% of the nation’s total population. The term “Hispanic,” or “Latino,” is applied to individuals sharing a common cultural heritage, most often in reference to a common language (i.e., Spanish). Although the term does not refer to race or common ancestry, Hispanics are typically treated as a single ethnic group despite having distinct sociocultural and genetic backgrounds. Genetically, researchers usually characterize Hispanics as a mix of Native American, European, and African ancestries. However, even this description does not fully capture the complex ethnohistories and populational migration patterns unique to the populations which fall under the umbrella term of Hispanic.

For instance, during the mid 19th- to early 20th-century, Chinese immigrants arrived in many Central and South American countries as indentured manual laborers. Following England’s abolition of slavery and its decline in the United States, the Chinese emerged as the major labor force for the sugarcane fields of Cuba and Peru. Concurrently, a community of ethnic Chinese formed in Panama around work on the Panama Canal Railroad Company. Despite the historical presence of Chinese populations in many Spanish-speaking countries, this biological variation is often overlooked in analyses of Hispanic skeletal remains. This study utilizes geometric morphometrics to examine regional variation among Hispanic populations in relation to Chinese affinity. Fifteen standard craniometric landmarks were collected from modern populations of Panamanians (n=22), Cubans (n=19), Peruvians (n=7), and Chinese (n=5) using a Microscribe™ G2X® digitizer in association with the software program *Three Skull*. Among-group variation was characterized using landmark-based Procrustes superimposition and thin-plate-spline approaches from geometric morphometry in conjunction with canonical variates analysis and Procrustes distances among groups. These analyses were performed in the shareware program MorphoJ. The first two canonical variates accounted for 98% of the total variation among groups. The Peruvian and Chinese crania clustered on opposite ends of a canonical variates plot, with the Cuban and Panamanian crania positioned between these two populations. Based on permutation tests (1,000 permutation rounds) for Procrustes distances among groups, all of the populations were significantly different from one another (p<0.05) in terms of shape variation. Procrustes distances were utilized to characterize group affinity due to the small sample sizes. The Chinese crania exhibited the most similarity with the Hispanic crania in the following order from most to least: Cuban (0.036), Panamanian (0.041); Peruvian (0.043). Overall, the most morphological similarity was found between the Cuban and Panamanian crania (0.0296), while the least was found between the Cuban and Peruvian crania (0.047). Graphical visualization of shape revealed varying patterns of shape change between the Chinese and Hispanic crania at lambda, prosthion, basion, opisthion, and asterion.

These results can be explained in terms of the unique

ethnohistories of Cuba, Panama, and Peru. Several morphological studies have shown Peruvians to be a relatively homogenous, indigenous population compared to other Latin American populations, such as Cuban and Panamanian which exhibit varying amounts of African and European admixture. The more heterogeneous nature of these two populations likely justifies their greater Chinese affinity compared to Peruvian crania. Not only does this study highlight the potential presence of East Asian ancestry in Hispanic populations, but reveals the importance of considering such information in standard forensic practice to create a more informative assessment of ancestry in unidentified Hispanics than is often utilized.

Ancestry, Hispanic, Geometric Morphometrics

H83 Stature Estimation Utilizing the Crural Index

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After attending this presentation, attendees will have a greater understanding of how body proportions, specifically the crural index, can be used as a categorical variable to group reference data in stature estimation models, which can be an effective alternative to population-based models.

This presentation will impact the forensic science community by introducing a new perspective on stature-estimation models which highlights the utility of body proportions as categorical grouping variables instead of race.

Stature estimation from an unknown individual is ideally derived from a model based on an appropriate reference population. In general, population-based stature-estimation models produce a more accurate point estimate, a narrow prediction interval, and a higher R2 value indicative of a tighter grouping of the reference data along the regression line. However, myriad circumstances exist where even population-based models either cannot be used in cases where ancestry cannot accurately be deduced or the model is derived from a sample that is antiquated, has an inadequate sample size, or does not accurately represent the diversity evident in the true population. Populations are ever-changing entities, both in terms of the volatility of their culturally based definitions as well as the susceptibility of their underlying genetic structure being influenced by stochastic events. As we become increasingly aware of these issues it is important to explore and evaluate the potential of other methods that have a more inherent biological foundation. This is the aim of the method proposed in this presentation, as it utilizes the crural index (tibia XLN/femur XLN) as a categorical variable in place of populations in regression analyses estimating stature from the maximum length of the femur (XLN). The crural index is often cited as an example of Allen’s rule and exhibits strong geographic patterning.

This research uses three sample populations, White (n=113), Black (n=52), and Chinese (n=48), with known statures and recorded femur XLN to create the regression equations for analysis. The White and Black sample populations were from a reference dataset for osteometric sorting created by Dr. Byrd and Dr. Adams.¹ The Chinese data is from Stevenson.²

Three different models were created to test the utility of the crural index as a grouping mechanism to better estimate stature. All of the models are based on grouping of individual samples based on their crural index, with the group boundaries determined three different ways, all based on the standard deviation of the sample population as a whole. The first model, MeanGr, contains two groups, one containing individuals with crural indices lower than the mean, and the other with indices greater than the mean. The

second model, CenterSDGr, designates three groups based on centering the standard deviation around the mean. The third model, MeanSDGr, contains four groups with the cutoff values being one standard deviation below the mean, the mean, and one standard deviation above the mean. Each crural index model was compared to the model containing all samples, as well as models for each population within the sample, representing the population-based stature methods that are widely used in forensic anthropology.

Descriptive statistics were produced for each model; the analysis of variance revealed all models were significant ($p < 0.001$). A regression analysis was conducted for each group within each model. The adjusted R² values for the population-based model ranged from 0.882 for the Black group to 0.638 for the Chinese group, with the White group at 0.765. The adjusted R² values for the MeanGr are 0.778 for Group 1 and 0.779 for Group 2. The adjusted R² values for the CenterSDGr are 0.815 for Group 1, 0.759 for Group 2, and 0.731 for Group 3. The adjusted R² values for the MeanSDGr are 0.872 for Group 1, 0.740 for Group 2, 0.809 for Group 3, and 0.693 for Group 4. Prediction intervals were also calculated in accordance with Giles and Klepinger, along with standard errors, as additional means to evaluate these models.³ This research demonstrates that stature-estimation models based on the crural index can be an effective alternative to population-based models.

References:

1. Byrd JE, Adams BJ. Osteometric sorting of commingled human remains. *J Forensic Sci* 2003;48:717-23.
2. Stevenson PH. On racial differences in stature long bone regression formulae, with special reference to stature reconstruction formulae for the Chinese. *Biometrika* 1929;21(1): 303-21.
3. Giles E, Klepinger LL. Confidence intervals for estimates based on linear regression in forensic anthropology. *J Forensic Sci* 1988;33(5):1218-22.

Stature Estimation, Crural Index, Human Variation

H84 Determination of Sex From the Human Hyoid Bone in a Contemporary White Population

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After attending this presentation, attendees will: (1) have an understanding of differences in accuracy rates of discriminant functions developed on White archaeological skeletal populations when applied to contemporary White skeletal populations; (2) have an understanding that methods developed on archaeological populations are not necessarily accurate for estimation of sex of modern populations; (3) learn how to apply discriminant function equations for estimation of sex from human remains; and, (4) learn how to test discriminant functions developed on archaeological populations on modern populations.

This presentation will impact the forensic science community by outlining the manner in which: (1) this is unique research that has not been published previously for a modern population; (2) this new data will help in the positive identification of unknown human skeletal remains in modern White populations; (3) this data shows that, for forensic cases, methods employed for estimation of sex must be developed using modern skeletal remains; (4) the *Daubert* and *Mohan* rulings require forensic methods to be scientifically tested and reproducible, which this

presentation does for the hyoid bone in modern White populations; and, (5) this research establishes methodologies for the hyoid bone in modern White populations via reproducible, scientifically tested results, in line with *Daubert* and *Mohan* rulings.

This project evaluates a method for sex determination using the human hyoid bone. It evaluates the accuracy of six discriminant functions developed by Kindschuh *et al.* on an archaeological white skeletal population and then applies the functions to a contemporary White skeletal population. The hyoid body and the left and right greater cornua were measured from 134 individuals (68 male; 66 female). Fifteen measurements were taken from fused hyoids and 12 measurements were taken from unfused hyoid bones. Applying discriminant functions developed from archaeological hyoid bones yielded accuracy rates ranging from 79.1% to 92.3% for contemporary White hyoid bones. Mean and sex specific accuracy rates indicated that two functions developed on archaeological fused hyoids were not accurate in determining females in a contemporary White skeletal population. Discriminant functions developed on the unfused hyoid and the hyoid body of fused and unfused hyoids had accuracy rates ranging from 88.1% to 92.3%, indicating that they were efficient for determining sex for a contemporary White skeletal population.

Two-sample *t*-tests showed statistically significant differences between archaeological and contemporary populations in the height of the anterior cornua (CHI) of both fused and unfused males. Significant differences were also observed between the archaeological and contemporary populations' Total Hyoid Length (THL) in both males and females. Four of the six discriminant functions developed by Kindschuh *et al.* can be applied to contemporary White hyoid bones; however, significant differences in THL and CHI between archaeological and contemporary skeletal populations indicate that discriminant functions developed solely on archaeological-fused hyoids are less accurate when applied to contemporary White hyoid bones.

The use of the hyoid bone for determination of sex from discriminant function calculations remains promising, with accuracy rates as high as 92.3% in contemporary White hyoids. In forensic contexts, this new data will aid in the positive identification of unknown human skeletal remains. The *Daubert* and *Mohan* rulings require forensic methods to be scientifically tested and reproducible. As secular and allometric changes have been observed in skeletal anatomy as populations evolve, it is therefore essential that methods developed on an archaeological population be tested for accuracy on a modern population.

Sex Determination, Hyoid, Modern

H85 Redefining Facial Soft Tissue Depth Landmarks for a Juvenile Australian Sub-Population: The Case of the Alar Curvature Point

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After attending this presentation, attendees will have been provided with an introduction to: (1) the error associated with the application of the current Alar Curvature point (AC) craniometric landmark definition; and, (2) the redefinition of this landmark for

juveniles in an Australian sub-population using 3D multi-slice Computed Tomography (CT) clinical data.

This presentation will impact the forensic science community by addressing the issue of the current AC landmark definition not being applicable to all demographics. It will introduce novel definitions for locating this landmark in juveniles from an Australian sub-population. It will also demonstrate the value of clinical CT scans as alternative sources of contemporary population-specific skeletal data.

A major objective in forensic anthropology is victim identification, which often requires access to antemortem records. Unfortunately, comparative methods that utilize antemortem records such as DNA and dental comparison are not always successful or available. In these instances, facial approximations have been successfully applied as an initial recognition tool. The most common facial approximation method uses published soft tissue thicknesses at various craniometric landmarks to "build" the face onto the skull. It is therefore important to ensure that these landmarks are located as reliably as possible. For the most part, craniometric landmark location definitions translate well when locating them on juveniles. However, definitions that include fixed metrical values would be limited to the age demographic from which they were developed. The only existing definition for the AC landmark is described as being approximately 3mm lateral to the border of the nasal aperture, which has been utilized for adults and juveniles alike.¹

As part of a larger study dealing with facial soft tissue thicknesses, this study aimed to test the error rate associated with the current definition of the AC landmark. In addition to this, it aimed to create novel definitions with lower error rates that could be utilized with a higher degree of confidence.

Forensic anthropology in Australia has been limited by a lack of documented juvenile skeletal collections; however, access to CT data in a clinical setting overcomes this. For this study, the sample comprises retrospective cranial CT scans (resolution: 0.5mm/0.3mm) of 21 juveniles aged from birth to 2 years. These were obtained from the Mater Children's Hospital, Brisbane.

Following the creation of 3D volume renderings of both bone and soft tissue, the Mid-Sagittal and Frankfurt Horizontal reference planes were constructed. The left and right AC landmarks were located on the skin surface according to the definition of being the most posterolateral position along the base of each nasal alar.² Points on the bony surface immediately posterior to the landmarks were added, and their position with relation to surrounding bony features was examined. Primarily, measurements from the Alare (Al) landmark on the lateral border of the nasal aperture to a parasagittal plane at the AC landmark ("X") were taken to assess whether or not there was significant difference from the suggested 3mm distance.

Preliminary analysis was conducted and no significant bilateral asymmetry was discovered ($p>0.05$). Consequently, measurements were considered for the left side only. Two-tailed Student *t*-tests indicate that all lateral measurements were significantly different from the current definition of 3mm, with a mean error of 2mm. A comparison of the measurements to the new mean (4.9mm±1.3mm) provided a smaller mean error of 0.6mm. "Y" measurements were also taken, to quantify the inferior position of AC in relation to Al (3.8mm±1.5mm). During this presentation, novel definitions of the AC location will be presented for the 0-10 years of age cohort. Overall, the AC landmark can be located with a higher degree of confidence for facial approximations.

References:

1. Stephan CN, Simpson EK. Facial soft tissue depths in craniofacial identification (part II): An analytical review of the published sub-adult data. *J Forensic Sci* 2008;53:1273-9.

2. Kolar JC, Salter EM. Craniofacial anthropometry: Practical measurement of the head and face for clinical, surgical and research use. Springfield: Charles C Thomas, 1997.

Facial Approximation, Craniometric Landmark, Computed Tomography

H86 Facial Soft Tissue Depth Shorths and Shormaxes for French Adults and the C-Table Data Store

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After attending this presentation, attendees will possess an appreciation for the utility that shorths and shormaxes hold to describe facial tissue depths in craniofacial identification, over and above the long-established arithmetic mean. The *TDStats R* package, specifically written for exploratory analysis of facial soft tissue depths and computation of shorths and shormaxes, will also be demonstrated.

This presentation will impact the forensic science community by providing a structured and systematic framework for face prediction that transcends quantification of the facial soft tissue thickness alone. Such a development is long overdue in the widely recognized subjective domain of facial reconstruction.

Humans show variation in soft tissue thickness at any given landmark of the face. However, it is impossible for smaller values to be, in relative terms, as extreme as the larger ones since soft tissue depths cannot be negative. Consequently, facial soft tissue thicknesses are likely to be positively skewed, especially around the chin and in the cheek regions where measurements can be large. This pulls the mean away from the densest regions of the data and reduces its descriptive value. Irrespectively, means have been used to describe facial soft tissue depth data for well over the past 100 years.

Recently, Stephan, Simpson, and Byrd suggested using shorths (the mean of the densest half of the data) to avoid this pitfall, and supplemented this statistic with another — the shormax (75th percentile between the shorth and the maximum) — to better approximate individuals who fell far into the right tail. This has the added benefit of utilizing the full spread of data in the right tail without invoking subjective groupings of body fatness and provides two point estimates. Using recently published facial soft tissue depth data collected on French adults, this study reports skew, shorths, and shormaxes in a contemporary sample measured with some of the latest technology. Positive skew in excess of 0.7 magnitude was found at 28% percent of 36 landmarks measured for French adults (n [max] = 469), and 31% for of 26 landmarks recorded in the C-Table (pooled data; n [max] = 756). Commonly skewed landmarks were rhinion, alare curvature point, mentolabial sulcus, menton, zygion, gonion, and mid-ramus. Differences between means and shorths at skewed landmarks were typically in the order of 1 to 2mm, while 75%-shormaxes were generally 2-3mm larger than the shorth down the midline, and up to 11mm larger at bilateral landmarks (e.g., gonion). This study confirms that some facial soft tissue depths landmarks, important for face prediction, are skewed and together with the fact that shorths provide identical results to means at normally distributed landmarks, highlights the broad-sweeping utility of shorths and shormaxes as central tendency descriptors for all craniofacial landmarks.

In forensic face prediction, shorths should be employed prior to shormaxes because they most closely approximate the densest half of the distribution. If advertisement of this first face

is unsuccessful, then a second face constructed using shormaxes should be undertaken. Such serial utilization of these statistics holds three strategic and key benefits: (1) it avoids cost and time expenditure on construction of rarer face types, unless there is a requirement for it; (2) it doubles media exposure, limiting chance oversight by relevant members of the public; and, (3) if the initial miss was underpinned by underestimation of the soft tissue depths, the second face holds the capacity to rectify the miss.

Whether the shorth/shormax combination generates smaller standard errors of the estimate overall compared to single-point estimates of regression equations is yet to be determined. However, the doubling of the media exposure provided by the dual-point estimates may be an unrivaled advantage (especially when body weight cannot be reliably predicted from hard tissues alone). Whatever the case, shorths and shormaxes provide significant improvements to the universally employed arithmetic mean.

Facial Approximation, Facial Reconstruction, Craniofacial Identification

H87 New Prospects in Facial Approximation: A Differential Approach in Mouth and Nose Reconstruction in Skeletal Classes

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After attending this presentation, attendees will understand some principles of mouth and nose reconstruction in facial approximation, a technique which strives to reproduce facial features and estimate the appearance of the antemortem face from human skeletal remains.

This presentation will impact the forensic science community by providing tools for a more precise reconstruction of nose and mouth from skeletal remains.

Nose: One hundred forty-two radiographs were randomly taken from diagnostic cephalometric X-ray films (lateral view) of Italian dental patients (aged from 14 to 47 years, with a mean age of 25) who consulted the Department of Orthodontics of Milan for dental treatment. Six measurements were taken: height of nasal aperture; nasal depth; nasal angulation; nasal-bone angle; nasal-bone length; and the projection of the previous segment to the line perpendicular to the Frankfurt Horizontal plane, in order to verify which one had the best correlation with nasal depth. For males, the height of nasal aperture had the best correlation with nasal depth; for females, the nasal-bone-projection had the best correlation with the nasal depth. Multiple regression was also conducted showing the need to distinguish the formulas for male and female nasal reconstruction, since they are significantly different. In this sample, the mean of the male nasal depth was 3.83cm (C.I.: 3.72cm–3.93cm); the mean of the female nasal depth was 3.42 (C.I.: 3.37cm–3.47cm). The difference between the two means is extremely significant (p-value = 0.0001), which means that nasal depth is deeply influenced by sex. The difference in nasal angulation between the two genders is significant and a novelty compared to previous studies. Moreover, the nasal depth was statistically different between Class II and III patients.

Mouth: At the present, lip and mouth reconstruction in different skeletal classes has been poorly investigated and this study focuses on emphasizing the importance of using different methods for selective reconstruction according to skeletal class. Sixty latero-lateral radiographs (twenty of each skeletal class) were randomly taken from diagnostic cephalometric X-ray films (lateral

view) of the same Italian sample.

The radiographs were divided into the three skeletal classes based on Riedel's ANB-angle, then the profiles were reconstructed with George's method. To evaluate the degree of reliability of George's reconstructions, engineers developed a new software, to assess the difference between the two areas (the real profile and the reconstruction), showing that the reliability of George's method is very low in pathological classes (II and III). To adjust his method to the different classes, seven new reference planes of reconstruction were identified, evaluated by the software and compared both with the real profile and with the classic George method. This comparison led to the evidence that Class III showed a highly statistically significant preferential plane to use for a better mouth reconstruction. Better planes were found also for Class I and II, but were not statistically significant. This work pointed out the basic importance of using different methods for nose and mouth reconstruction by differentiating between males and females and the three skeletal classes.

Facial Reconstruction, Nose, Mouth

H88 Cognitive Bias in Forensic Anthropology: Visual Assessments of Skeletal Remains Are Susceptible to Confirmation Bias

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After attending this presentation, attendees will understand how contextual information can influence forensic anthropologists' judgments and decision making in non-metric assessments.

This presentation will impact the forensic science community by demonstrating that cognitive bias can alter and impact forensic anthropology and increase awareness of how contextual information can affect objective assessments, interpretations, and final conclusions of forensic anthropologists.

The issues of cognitive bias and its effects in forensic science and criminal investigations have been increasingly discussed and described. The National Academy of Sciences in the United States and the Forensic Regulator in the United Kingdom have highlighted the review of standards and processes within forensic science disciplines and underlined the potential for bias in forensic interpretations. Studies have demonstrated the effect of cognitive bias in decision making within a number of forensic fields. Research has shown that judgment and decision making can be influenced by extraneous information that can lead to a decrease of objectivity in the interpretation of forensic evidence. In many disciplines, such as forensic anthropology, the presence of cognitive interpretation issues and its impact is still not fully assessed with a lack of empirical studies that test the degree to which cognitive issues might arise and affect the judgment and final evaluation of the forensic anthropologist. The anthropological methods are acknowledged for being subjective (in a similar manner to many other forensic science disciplines), hence the need for research in this area. This study investigated if confirmation bias can arise in visual perceptual assessments within biological profiling on skeletal remains when subjected to different contextual information.

An experimental study was designed to examine cognitive biases within forensic anthropological non-metric methods in assessing sex, ancestry, and age-at-death. To examine viewer interpretation, 41 participants were asked to establish

a full biological profile on skeletal remains of one complete individual. The skeletal remains had ambiguous features, with the morphological traits of the skull and pelvis showing no clear signs of female or male characteristics. This was particularly significant in this study as cognitive biases are more prevalent in ambiguous cases. Participants were semi-randomly divided into three groups. Two of the groups were given contextual information regarding the sex, ancestry and age-at-death of the individual before conducting the analysis. The third group acted as a control group with no contextual information provided prior to the assessment of the same skeletal remains. The experiment was designed to investigate if the interpretation and conclusions of the skeletal remains would differ among participants within the three groups when exposed to contextual information and to assess if the examiners would confirm or disagree with the given context when establishing a biological profile.

The results revealed a significant biasing effect within the three groups, demonstrating a strong confirmation bias within participants' assessments of sex, ancestry, and age-at-death. In assessment of sex, the control group was divided with 31% assessing the remains to be male and 69% female. However, in the group that received contextual information that the remains were male, 72% indicated the remains were male, 14% female, and 14% undetermined in their conclusion. Of the group that was given the context that the remains were of a female, 100% of the participants concluded the remains to be female. Similar results were obtained for ancestry and age-at-death assessments. The results demonstrated that participants given contextual information before conducting the analysis had a strong bias to conform with the given context when conducting the examination. This study demonstrates that cognitive bias exists in forensic anthropological non-metric methods on skeletal remains and affects the interpretation and conclusions of the forensic scientists. The presentation will present the findings and discuss the importance of recognizing biases that may impact interpretation during analysis as well as highlighting the need for further research addressing how to minimize and alleviate these effects to secure a more robust and credible discipline.

Forensic Anthropology, Cognitive Bias, Decision Making

H89 Enough is Enough! What Are Validation Studies of Age Estimation Methods Really Telling Us?

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After attending this presentation, attendees will learn about the relevance and construction of age estimation method validation studies and method development with respect to design (phase versus component systems) and statistical considerations.

This presentation will impact the forensic science community by offering suggestions for research on age estimation methods and by demonstrating the need for revising phase systems with component systems.

The literature is flooded with validation studies of various aging methods on different populations. Because of the relative ease and speed with which these studies can be completed, they are ideal student research projects or conference papers and are generally viewed as valuable contributions to the research literature. The present study developed in similar fashion. The primary intent was to test the applicability of the Suchey-Brooks (SB)

pubic symphysis method on the modern Colombian population. A secondary goal was to assess whether any features were difficult for a non-native English speaker with limited experience using the system to evaluate (i.e., to gauge whether the definitions and features translated well into Spanish or if any were "English-centric"). To this end, the lead researcher deconstructed the SB system into components by selecting eight key features from the phase descriptions and assigning states of expression to each feature based on the SB descriptions of their age progression morphology.

The following features and states of expression (in parentheses) were scored as components: pubic tubercle (absent, continuous with symphyseal face, separate); symphyseal face topography (billowed, flat, depressed); ventral rampart (absent, partial, complete, deteriorating); dorsal rim (absent, partial, complete, deteriorating); porosity (absent, present); lipping (absent, present on dorsal margin, present on ventral margin, present on both margins); oval outline (incomplete, complete); and, bony ligamentous outgrowths (absent, present). In addition, each pubic symphysis was assigned an SB phase. The sample consisted of right and left pubic symphyses from individuals in the Colombian Modern Skeletal Collection at the Institute of Legal Medicine in Bogota, Colombia (n=60, mean age=45.6 years, SD=23.1, range=24-93). Each observer had >5 years experience with pubic symphyseal aging, though the native English-speaking observer (Observer 1) had more experience with the SB system than the Spanish-speaking observer (Observer 2). Reference casts were used, and the definitions were translated into Spanish by a professional interpreter. Cohen's Kappa coefficients were calculated (SPSS® Version 20) to examine observer agreement and to evaluate whether certain traits were difficult to assess. In addition, a linear regression was run with "phase" as the independent variable, "age" as the dependent variable, and accuracy rates were calculated for each observer's scores.

Phase scores recorded by Observer 1 correctly estimated age in 93% of the sample using the 95% age ranges in the SB system; the corresponding accuracy rate for Observer 2 was 87%. The linear regression R² values were 0.64 and 0.49 for Observer 1 and 2, respectively (R=0.80 and 0.70, with no significant difference between correlation coefficients (z=0.834, p=0.404)). Observer agreement was relatively low for the SB phases (0.47), but ranged from relatively strong to very strong for the components (0.71 - 0.98). The component features ranked from highest to lowest Kappa are bony ligamentous outgrowths (0.98), oval outline (0.89), porosity (0.87), symphyseal face topography (0.86), ventral rampart (0.84), dorsal rim (0.80), pubic tubercle (0.75), and lipping (0.71).

The accuracy rates indicate that the SB system is adequate for use on the modern Colombian population. The slight discrepancy between observers could be attributed to experience level and/or language/translation issues. However, the high agreement for individual components indicates that the discrepancies are unlikely due to language problems. Instead, the comparatively low agreement for phase assignment indicates that a phase system is more difficult to apply objectively. With phase scoring, error associated with the individual traits is compounded, there are no suggestions on how to weight traits or how to handle variation from the typical Gestalt, and thus deciding between two phases is often subjective. The Kappa values also indicate that coding possibilities for each component should not exceed two or three states of expression, each with minimal overlap. It is anticipated that component-based methods will reduce observer error and generate more accurate age estimates. However, the soundest age estimates will be derived from multifactorial methods developed using probability statistics and Bayesian estimation. Seasoned and burgeoning researchers are encouraged to explore component systems in future research on skeletal age estimation.

H90 Palate Shape and Depth: A Shape-Matching and Machine-Learning Method for Estimating Ancestry From Skeletal Remains

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After attending this presentation, attendees will have learned the efficacy of palate shape as a skeletal ancestry indicator, the "typical" shape of the palate for individuals of Hispanic ancestry, the potential usefulness of palate depth in assessing ancestry from the human skeleton, and the influence that secular change may have on traditional indicators of ancestry.

The presentation will impact the forensic science community by demonstrating the usefulness of this technique in assessing the ancestry of an unidentified individual. The ultimate goal of forensic anthropology is to identify the dead. This new technique adds to the catalog of techniques available in that pursuit and provides new information regarding the population-based variation of the human skeleton.

The assessment of ancestry from skeletal remains is a vital aspect of the biological profile so often used in forensic anthropology. As such, a plethora of techniques has been developed to examine this aspect of the biological profile. Gross observation of morphological features and reliance on observer experience to correctly interpret those features are traditional methods of analysis. However, as replicability has become more important in both the laboratory and the courtroom, objective, metric techniques of analysis have become increasingly prominent. This study attempts to merge these two areas: by taking a feature that is traditionally examined non-metrically, palate shape, and, by using computer-generated models, evaluate it quantitatively.

Using 3D digitizer technology in conjunction with shape-matching and machine-learning methods common in computer science, palate shape curves were collected from 376 individuals of varying backgrounds from both historic and modern contexts. Additionally, the digitizer was used to capture palate depth (superior to inferior), which is a novel measurement in this study. The crania used in this study came from the Pima County Office of the Medical Examiner (PCOME) in Tucson, Arizona, the William M. Bass Donated Skeletal Collection at the University of Tennessee, Knoxville, and the Roger J. Terry Skeletal Collection, the Smithsonian Institution's National Museum of Natural History in Washington, D.C.

Computer analysis of the data indicated that palate shape was an accurate indicator of ancestry in the whole sample 58% of the time, which is higher than would be expected from random chance (33%). This number improved slightly when the historic sample was examined on its own (61%). Cluster analysis of the curves revealed that the parabolic, hyperbolic, and elliptical shapes, and by extension the White, Black, and Asian/Native American ancestry groups, are relatively discrete from one another. The only significant overlap in shape was observed between White and Hispanic individuals, both being characterized by a parabolic palate shape.

The results of the depth measurement in this study are preliminary at this stage. However, some interesting findings came to light. Measurements indicated that, at the intersection of the

transverse palatine suture and the median palatine suture, palate depth differed among the ancestry groups, with individuals of Hispanic ancestry having the deepest palates.

This study has resulted in several important conclusions. First, the traditional methods of ancestry estimation should be quantified, so that they may continue to be used to identify the unknown in arenas where objective measures are necessary. Second, these results demonstrate that the skeletal anatomy of Hispanic individuals, especially in regard to ancestry, varies in unexpected ways, and warrants further research. Finally, the preliminary results presented here indicate that examining palate depth may be an important tool in the future of the discipline.

Palate Shape, Ancestry, 3D Digitizer

H91 The Effects of Parturition on the Areas of Interest for Age Assessment on the Pelvis

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After attending this presentation, attendees will understand the need for consideration of parity when assessing the age of the pelvis in a recent forensic sample from the United States, the necessary methods utilized on the pelvis, the process and reasoning behind the collection of data, and subsequent statistical analysis employed in this study.

This presentation will impact the forensic science community by indicating whether parturition effects an accurate assessment of age on the pelvis in a recent forensic sample from the United States and if it needs to be considered as an addition to the methods employed in human identification.

Assessing the age of an adult skeleton is a critical facet in creating the biological profile of an individual. To date, there are different methods used on select elements of the skeleton to assess the age of the individual and, currently, the pelvis is relied upon heavily to obtain accurate and reliable age ranges.¹⁻³ Many have stated that age-related changes follow different trends in males and females, with parity presented as one of the possible causes for such differences.^{1,4-6} There is reason to believe that parturition may increase the rate at which the areas of interest of the pelvis degenerate. However, this hypothesis has yet to be formally tested on a recent skeletal collection. The purpose of this study is to assess the effects of parturition on the pubic symphysis and auricular surface and determine whether it influences the physiological age of the individual enough to cause an inaccurate estimate of the chronological age.

Data were collected from the William M. Bass Skeletal Collection located at The University of Tennessee, Knoxville. This is a collection of recent forensic skeletons with known age-at-death, ancestry, sex, and medical background. The areas of interest are the pubic symphysis and the auricular surface and were analyzed in accordance with the methods included within Buikstra and Ubelaker's Standards For Data Collection From Human Skeletal Remains as well as methods proposed by Buckberry and Chamberlain and Berg.^{1,3,7} Therefore, others attempting to replicate this research will be able to reliably assess these areas. Time was designated at the beginning of the second and third days to employ the test-retest method to calculate the intra-observer error rate and ensure reliability of these assessments.

In this study, a statistical comparison was made between females who have given birth and those who have not to determine whether this process affects the rate of degeneration of the areas of interest of the pelvis. A simple linear regression was run to determine the prediction ability of the accurate phase of the pubic symphysis and auricular surface from the individual's age. This was performed for each method of analysis. The results were then

compared between the parous and nulliparous groups. In addition, the individuals who were aged incorrectly were analyzed to determine whether there was a pattern of over-estimation unique to the parous group and, if so, whether the over-estimation is enough to cause an inaccurate assessment of the chronological age of the individual.

The study contained 434 individuals (234 males, 200 females, 157 parous, and 43 nulliparous). The data was entered into SPSS computer program version 20.0. A simple linear regression analysis produced significantly different results between parous and nulliparous females using the pubic symphysis but not the auricular surface. The current research suggests that parturition affects the pubic symphysis and not the auricular surface when determining age-at-death at the 95% confidence level. There is some factor that causes the parous females to appear, on average, ~1.5 years older than nulliparous females; however, the slopes of the regression lines indicate that the parous females age more slowly than nulliparous females, who are, on average, aging 2.67 times faster. An analysis of the incorrectly assessed individuals shows that parous females tend to be over-aged when compared to nulliparous females. The applicability of taking parturition into consideration when assessing the age of females for use in human identification in modern forensic cases will be discussed.

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Forensic Anthropology, Age Estimation, Pelvis

H92 Morphoscopic Trait Frequencies of Southeast Asians and Pacific Islanders

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After attending this presentation, attendees will understand the frequency distribution of morphoscopic traits among populations of the Pacific Islands and regions of Southeast Asia, the biological relationships between the populations of these regions with regard to those traits, and where these populations fall in a forensic ancestry classification.

This presentation will impact the forensic science community by providing data on the distribution of morphoscopic traits among populations of various islands of the Pacific and

oceanic regions of Southeast Asia and illustrate the variability between each region. This presentation will also demonstrate how individuals from these regions are categorized in comparison to individuals from mainland Asia as well as individuals of European, African, and Native American ancestry for a forensic application.

When assessing ancestry in a forensic context, individuals are generally classified into one of four categories: belonging to European, African, Asian, or Native American ancestry. This leaves out many populations that are generally more difficult to assess due to ancestral admixture and/or limited research about the population in question. With only these four ancestry assessments, individuals from Southeast Asia and the Pacific Islands are usually phenotypically classified as Asian. While the oceanic regions of Southeast Asia and the Pacific Islands will undoubtedly have morphoscopic trait frequencies similar to those of mainland Asia because of their shared ancestral lineages, there is still a great deal of variability in this region with genetic drift being an important factor. Previous studies of this region using craniometric data have shown a separation between Southeast Asia and North/East Asia, while also noting a close connection between island Southeast Asia and mainland Southeast Asia.^{1,2} Inter-regional clinal patterns of variation in these regions have also been noted by previous researchers using both craniometric and non-metric data.^{1,3} This provides evidence for a possible distinction between morphometric trait frequencies of populations of the oceanic region and those of mainland Asia.

To address the variability within these regions and where they fall in a forensic classification, data were taken using Osteoware™ on sixteen morphoscopic traits: ANS; INA; IOB; MT; NAS; NAW; NBC; NBS; NO; NFS; OS; PBD; PZT; SNS; ZYC; and, TPS.⁴ Skeletal specimens include (n=150) from the physical anthropology skeletal collection at the Smithsonian National Museum of Natural History and (n=33) from the University of Pennsylvania Museum of Archaeology and Anthropology's physical anthropology skeletal collection, for a total sample of (n=183). Populations collected include individuals from Indonesia, Malaysia, New Zealand, French Polynesia, Papua New Guinea, Fiji, and the Philippines.

A separation between groups can be clearly seen when morphoscopic trait frequencies of the Asiatic populations are compared with trait frequencies of European and African groups. As the different Asiatic groupings are narrowed down, a separation is still present but with more ambiguity. Canonical variance and principal component analyses illustrate that the island Southeast Asian and Pacific Island populations can be slightly differentiated from the mainland Asian population, though not with great clarity. When the island Southeast Asian and Pacific Islander populations are incorporated into the Asian groupings for frequency distributions, a much higher within-group variance is found, indicating that groups from these two regions may not work well within the larger Asian grouping and could potentially be separated from the larger Asiatic grouping. In conclusion, most individuals from Southeast Asia and the Pacific Islands tend to fall near the Asian categorization, though there is a slight separation from the mainland Asiatic population suggesting differentiation between mainland Asians and Southeast Asians/Pacific Islanders may perhaps be possible for a forensic ancestry assessment.

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Ancestry, Morphoscopic Traits, Pacific Islands

H93 Secular Change in Morphological Pelvic Traits Used for Sex Estimation

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After attending this presentation, attendees will learn of the secular changes occurring in three popularly used morphological traits of the pubis bone in forensic anthropology for sex estimation.

This presentation will impact the forensic science community by informing attendees about the variation present in trait expression and how these traits have changed through time. Furthermore, the implications for sex estimation methods using these traits will be discussed.

Phenice's technique using three traits of the pubis — the ventral arc, subpubic concavity, and medial aspect of the ischio-pubic ramus — is overwhelmingly the most preferred method of morphological sex estimation in forensic and biological anthropology.^{1,2} The original method was created using a historical sample of individuals born in the late 19th and early 20th centuries, as was a subsequent revision of the method including statistical analyses.^{3,4} Size and shape changes in the pelvis, and specifically the innominates, between historical and modern samples have been documented using metrics and geometric morphometrics; therefore, morphological trait expressions may also be changing through time.⁵⁻⁷

The purpose of this research was to evaluate changes in the expression of the Ventral Arc (VA), the Subpubic Sontour (SPC), and the Medial Aspect of the ischio-pubic ramus (MA), as described by Klales *et al.* by comparing trait scores from a sample of innominates from the Hamann-Todd Osteological Collection (HTH / historical) and the W.M. Bass Donated Collection (UTK / modern).⁴ The sample consisted of 136 females (HTH n=83, UTK n=53) and 163 males (HTH n=87, UTK n=76). Each individual was scored on an ordinal scale from one to five for each of the three Phenice traits using the Klales *et al.* method.⁴ Previous research revealed no significant differences in trait expression between ancestry groups (Blacks and Whites); therefore, ancestry groups were pooled for each sex, in each temporal sample for all further analyses.⁴

A Fisher Freeman-Halton Exact Test was run to determine if significant differences existed between the two temporal samples for each of the three traits by sex. Significant differences were found for all three traits in females: MA $p=0.004$; SPC $p<0.001$; and VA $p<0.001$. In males significant differences were found in two of the three traits between the HTH and UTK samples: MA $p=0.102$, SPC $p=0.002$, VA $p=0.008$. To determine which specific trait scores produced significant differences between temporal periods, the residual for each trait score was converted into a z-score and compared to the critical value. Analysis of the residuals for the medial aspect revealed greater-than-expected "ultra-feminine" expression (i.e., narrow ischio-pubic ramus with sharp ridge of bone present / score 1) in the modern sample females. Similarly in the modern sample, "ultra-feminine" expression of the subpubic concavity (i.e., well-developed concavity/score 1) and of the ventral arc (i.e., angled arc present with triangular portion of bone inferiorly/score 1) were greater than expected, while the historical sample had lower-than-expected residuals for the same scores. In males, the historical sample had a higher-than-expected "ultra-masculine"

expression of the SPC (i.e., large convexity/score 5), while in the modern sample this same score was lower than expected.

Analysis of the residuals reveals a trend toward more gracile females in the modern sample based on greater extreme low score trait expression. In all three traits, the modern females show greater-than-expected "ultra-feminine" expression (scores of 1 for each), while in historical males, only the subpubic concavity (score 5 only) had a residual above the critical value. A morphologically more "feminine" appearing pubis region parallels the metric data that shows an increase through time in pubis length, giving the region a more "stretched" appearance.⁷ The metric findings likely correspond to a narrower ischio-pubic ramus, a wider area lateral to the symphyseal face, and a greater subpubic concavity. Given the trends, it is apparent that secular change is occurring in morphological traits of the pubis, specifically with morphological expressions associated with females becoming more prevalent through time. Presumably, sex estimation in modern individuals using these traits and the aforementioned methods should have greater differentiation between males and females, with females classifying better given the greater extremes in gracile scores.

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Phenice, Pelvis, Non-Metrics

H94 Sex Determination From the Greater Sciatic Notch of Koreans Using 3D Models

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The goal of this presentation is to report sex determination

of the Korean population using 3D models. Applying the metric method to 3D bone models reconstructed from computer tomography scans, this presentation will illustrate the high accuracy of sex determination of this method.

This presentation will impact the forensic science community by explaining that sex determination from 3D models is a relatively new method of forensic anthropology. Using 3D bone models for the metric method proved higher accuracy accompanying objectivity when compared to former studies.

Using the pelvis is the most preferred method for determining sex in forensic anthropology. It is known that the accuracy of sex estimation is the highest when utilizing the pelvic bones because there are differences of function and structure of the pelvis according to the sex. Among numerous indicators, the greater sciatic notch is usually selected for the time of visual observation. This method has a few merits of taking less time and easy application on the dry bone in the field, but has the shortcoming of dependence on the accuracy on the observer's experience or subjectivity. As the importance of repeatability of a method is considerable, the visual analysis method is suspected of being inaccurate and inapplicable for the court issues. Therefore, this study developed a method to objectively measure three-dimensional models for sex estimation of the greater sciatic notch.

Studied were 103 sides (52 female sides, 51 male sides) of the Digital Korean Database stored in the Catholic Institute for Applied Anatomy. After defining the position of the Posterior Inferior Iliac Spine (PIIS) and the Ischial Spine (IS), the most medial point from the plane which passes the two points (PISS and IS) was defined as the Anterior Point (AP). The point where a line connects the PIIS and the IS and a line drawn from the AP meets vertically was defined as Posterior Point (PP). Mimics® version 15 was used to create 3D models and measure them with a measurement template designed for this study. There were 11 measurements related to distances between points and angles. For the sex determination process, SPSS version 20.0 was used to enable discriminant analysis.

The discriminant analysis on univariate data showed the range of accuracy from 53.8% to 96.2%. The angles of IS-PIIS-AP, PIIS-AP-IS, and especially, PIIS-AP-PP showed the highest accuracy. Compared to the visual examination of a former study, sex estimation with 3D model measurement possessed objectivity and accuracy.

Sex Determination, Sciatic Notch, 3D Model

H95 Sex Determination From Tarsal Dimensions

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After attending this presentation, attendees will understand the benefit of using tarsal dimensions for differentiating sex in European Americans.

This presentation will impact the forensic science community by outlining the results of a validation study using tarsal dimensions to differentiate sex in modern European Americans. This presentation will outline the most successful equations for estimating sex, as well as introduce error rates for use in forensic contexts.

Sex differentiation is an integral aspect of the biological profile. When available, the pelvis and cranium are highly successful at sex differentiation. However, postcranial metric analyses provide valuable substitutes when the pelvis or cranium is unavailable. In 2012, Harris and Case published a sex differentiation method using

dimensions from all seven tarsals of modern European Americans.¹ The study presented 42 equations (univariate and multivariate) for 18 separate measurements of the tarsals with accuracies as high as 93.6%. The study documented significant asymmetry in both male and female tarsal dimensions, and thus presented separate equations for the left and right sides, with the right side dimensions having the highest reported accuracies.

The current study proposed to test the Harris and Case equations on a modern European American sample from the Texas State Donated Skeletal Collection housed at the Forensic Anthropology Center at Texas State University-San Marcos (FACTS) in order to fulfill the best practices guidelines as outlined by the Scientific Working Group for Forensic Anthropology (SWGANTH). The sample (N=41) consists of 24 adult males and 17 adult females ranging in age from 19 to 102. The measurements were taken following Harris and Case using a standard osteometric board. A small subsample (n=5) was measured twice by a single researcher and by two additional researchers to evaluate intra- and interobserver error. Accuracy was calculated for males, females, and the pooled sample as a percentage based on the number of correct classifications for each equation on the left and right sides. Observer error was calculated using the measure of Mean Absolute Difference (MAD).

The results show accuracy rates ranging from 59-90% in the pooled sample, 58-100% in the female sample, and 52-96% in the male sample. For the pooled sample, the highest accuracy (90%) was calculated from the univariate equation for talus length from the right side. For the female sample, the highest accuracies (100%) were calculated from the right side for the univariate equations using talus height and cuneiform I length, as well as the multivariate equation using the cuneiform 1. For the male sample, the highest accuracy (96%) was calculated for the univariate equations based on talus length. The intra- and inter-observer error MAD measures ranged from 0 to 0.70mm.

While the resultant accuracy rates did not always agree with those previously reported, the original pattern of the right side equations performing better than those based on the left side did remain. In agreement with the Harris and Case study, the talus measurements did perform best on this sample. Among the univariate equations of the pooled sample, the talus length correctly classified each individual with the highest level of accuracy (91%) followed by the talus height. For the whole bone equations of the pooled sample, the cuneiform 1 from the right side correctly classified each individual with the highest accuracy (87%). For the equations that used multiple tarsals, the equation that used cuneiforms 1, 2, and 3 classified each individual correctly with the highest accuracy (87%). In addition, the observer error rates were minimal, suggesting high repeatability of the described measurements.

Overall, the equations performed well on the sample. The high levels of accuracy achieved (over 90%) for the pooled sample suggest that this is a viable approach to sex differentiation in European Americans when differential preservation or recovery limits the availability of skeletal material for analysis. This presentation will also discuss the difficulties associated with tarsal measurements, the effects of instrument choice for metric analysis, and the application of the equations to individuals of other biological races.

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Sex Estimation, Tarsals, Metrics

H96 Categorization of Human Crania Using Cranial Indices in a South Indian Population

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After attending this presentation, attendees will be able to recognize the details and significance of categorization of human crania using cranial indices in a South Indian population.

This presentation will impact the forensic science community by recognizing the possibility of categorization of human crania by cranial indices in a South Indian population which may be an important component in identification of highly decomposed dead bodies and skeletal remains.

Study of the human cranium is a common practice in physical anthropology that has drawn the interest of researchers. The human skull has been the most extensively studied bone for establishing the taxonomies at evolutionary levels by physical anthropologists. Crania are also the most commonly used skeletal elements in population studies because they are known to be more genetically driven and less affected by environmental factors. Due to lack of traditional skeletal collections like the Raymond Dart collection, Todd and Lyon repository, William M. Bass donated collection, and many more available in the Western world, only limited studies are available on Indian crania. Hence, it becomes difficult to make forensic standards for various population groups residing in India. Forensic scientists require population-specific data banks for identification purposes. India is a vast country having many ethnicities and population groups. To make population-specific forensic data banks for identifying individuals, there is a need for conducting investigations regarding human and population variations. The present study is an attempt to develop population-specific classification of crania using cranial indices that may assist the forensic anthropologists in the categorization of human skulls.

The sample for the present research included 118 dry adult crania that were obtained from a teaching institution in South India. All the osteometric measurements were taken using standard anthropometric instruments, and three indices, namely cranial index, orbital index and index of foreman magnum, were calculated. Cranial Index (CI) is calculated as (maximum cranial breadth/maximum cranial length) X 100, Orbital Index (OI) as (orbital height/orbital breadth) X 100, and Index of Foreman Magnum (IFM) as (Antero-posterior diameter/Transverse diameter) X 100. The crania were further classified based on these indices. The cranial index ranged between 66.67 and 85.71 (Mean \pm SD=78.57 \pm 4.11), the orbital index ranged between 68.89 and 102.63 (Mean \pm SD=84.23 \pm 6.64), and the index of foramen magnum ranged between 68.57 and 96.88 (Mean \pm SD=79.71 \pm 6.98). Cranial index did not show any significant correlation with the orbital index ($r=-0.162$, $P=0.081$) or the index of foramen magnum ($r=-0.045$, $P=0.626$). A statistically significant correlation was; however, observed between orbital index and index of foramen magnum ($r=-0.232$, $P=0.012$). The present study has attempted to classify the skulls based on the index of foramen magnum possibly for the first time. The present study has thus developed population-specific classification of crania using cranial indices that may assist forensic anthropologists in the categorization of human skulls. This categorization may be an important component in identification of highly decomposed dead bodies and skeletal remains. More such studies need to be conducted to understand the effect of environment and genetics on cranial shapes in different population groups.

Human Cranium, Cranial Indices, South India

H97 Sex Estimation Using Metric Analysis of the Human Clavicle

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After attending this presentation, attendees will have learned the degree of sexual dimorphism present in the human clavicle and several discriminant functions useful for the estimation of sex from both whole and fragmentary clavicles.

This presentation will impact the forensic science community by improving practitioners' ability to estimate the sex of skeletonized human remains when pelvic and cranial remains are not available or are too taphonomically damaged for analysis.

The development of methods for sex estimation using skeletonized postcranial remains other than the pelvis and cranial elements is imperative for physical anthropology to improve the reliability of biological profile estimates in cases of incomplete and/or fragmentary skeletal remains. As one of the last skeletal elements to complete fusion, the clavicle has the most extended period of post-pubertal development, making it an ideal element for use in sex estimation. Previous metric studies of the clavicle used the clavicle in combination with other skeletal elements to estimate sex with varying degrees of accuracy (Thieme specific accuracy not given and Frutos 92%).^{1,2} Previous research using only the clavicle to estimate sex produced limited success (60-72% accuracy), although the sample population (Indian) is not demonstrably sexually dimorphic.³ The use of computer-automated measurements improves the ability to estimate sex using the clavicle (92% accuracy), although the applicability to field work is limited due to the expense and time required for the use of computer-automated technology.⁴ The Hamann-Todd Collection was chosen because it could yield a greater sample size than most modern collections, thus developing a stronger method prior to adjustment for secular change in modern populations.

The three standard measurements of the clavicle (maximum length, sagittal diameter at midshaft, and vertical diameter at midshaft) were recorded along with 15 measurements developed for this study. The developed measurements include circumferences, lengths, widths, and angle heights, derived to decipher and record the complex pattern of sexual dimorphism in the clavicle. Sexual dimorphism in the clavicle was assessed using these 18 measurements of the left clavicle of 265 (132 females; 133 males) adults (fully fused clavicles) from the Hamann-Todd Collection. Independent samples *t*-tests with Bonferroni correction show male and female means differ at a statistically significant level for all 18 variables ($p<0.05$). A discriminate function analyses using the stepwise method (0.05 to enter, 0.10 to exit) produced a four-variable model with cross-validated accuracy of 89.8%. The measurements used in the four-variable model include the maximum length, circumference at midshaft, the height of the acromial end, and the sternal end width. A holdout sample from the Hamann-Todd Collection ($n=30$) similar in demographic character to the calibration sample was tested using the four-variable model. The accuracy of the four variable model on the holdout sample was 90.0%. Additionally, a three-variable, a two-variable, and four single-variable models were developed for use with fragmentary and taphonomically damaged remains. They also have high predictive power (75.1-88.3% cross-validated calibration sample; 60.0-93.3% hold-out sample). This study suggests the sternal end has more predictive power than the acromial, which likely corresponds to the later fusion of the sternal end. Circumference and length are also highly predictive, as they are used in all multivariate models along with being useful on their own.

This method serves to supplement, not replace, sex estimation based on the traditionally highly accurate pelvic and cranial remains. Further research is required (and planned) to confirm these methods are applicable to modern populations.

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Sex Estimation, Clavicle, Metric Analysis

H98 Sex Estimation Using Complete and Fragmentary Cuboid Bones

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After attending this presentation, attendees will understand how the cuboid bone can be used to aid in the estimation of sex of unknown individuals.

This presentation will impact the forensic science community by providing an additional means for sex estimation of incomplete and fragmentary skeletal remains.

Sex estimation is a vital step in the correct identification of unknown individuals. Expanding sexing methods to include unused and under-used bones will increase the accuracy of this assessment, especially in cases where the remains are incomplete or fragmentary. In 1976, Steele was one of the first researchers to examine sexual dimorphism of the talus and calcaneus.¹ Since then, other researchers including Barrett *et al.*, Bidmos and Asala, Bidmos and Dayal, Gualdi-Russo, Murphy, and Wilbur have repeated Steele's research and established techniques based upon the talus and calcaneus as useful tools in sex estimation with accuracies as high as 96%.²⁻¹⁰ Additionally, they have shown that these techniques can be applied to various populations from both the past and present. However, little research has been done on the remaining five tarsals. In 2009, using the William M. Bass Skeletal Collection and a mini-osteometric board, Sheena Harris measured the maximum length and width of all seven tarsals.¹¹ Her measurements required the bones to be in their complete form and the study did not examine the smaller segments of the bones. In 2011, Schmuhl demonstrated that the three cuneiforms, regardless of their completeness, can be just as useful in sex estimation.¹² These results indicate that other tarsals may be of value as tools for sex estimation.

This study looks at the cuboid from 100 adult skeletons (50 male and 50 female) from the William M. Bass Skeletal Collection for their usefulness in sex estimation. The study first examines the maximum length that was used by Harris from complete cuboid bones and then examined additional original measurements (such as specific articular surfaces) that divide the bone into smaller segments.¹¹ Digital sliding calipers were used to take 12 measurements (1 following Harris and 11 new measurements) of the left cuboid.¹¹

FORDISC® 3.0, was used to perform Discriminant Function Analysis (DFA) to test multiple measurements for their ability to discriminate by sex.¹³ With an overall accuracy of 86.8%, this study

shows that the cuboid has value as a sexing instrument. This study further shows that only a small portion of the cuboid needs to be present to be useful. Following the baseline suggested by Scheuer and Elkington, measurements (univariate) with accuracies equal or greater than 80% were considered to be useful in this study.¹⁴ Five out of the 12 univariate measurements taken in this study meet or exceed that baseline. When various multivariate approaches are taken into account, the accuracy of the cuboid rises even higher. While some of the individual variable accuracy rates fell below 80%, that rate increased above 80% when combined with at least one other measurement. Another multivariate approach taken into account involves relative weight. For instance, when all 12 measurements are used in a DFA, the overall accuracy rate is 86.8%. However, if only the measurements with the highest relative weights of these 12 measurements are considered and the lowest relative weights are dropped, the accuracy rises to 93.5%. In conclusion, although the cuboid is one of the smaller bones of the body, it should be considered a useful tool for estimating sex regardless of its completeness.

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Cuboid, Dimorphism, Sex

H99 Estimation of Cartilaginous and Soft Tissue Components for Estimating Adult Stature Using the Anatomical Method

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The goal of this presentation is to present an improved method, by using a digitizer, for estimating skeletal height using the standing anatomical position. Once accurate individual skeletal heights are obtained, regression formulas that account for the Cartilaginous and Soft Tissue (CCST) height components are used to estimate biological statures.

This presentation will impact the forensic science community by demonstrating how the CCST is related to age and individual variation. This presentation will discuss how to improve the accuracy of the anatomical method of stature estimation and how to create multiple stature-estimation formulas for skeletons where age-at-death is known but antemortem stature records are lacking.

Thirty-eight European American males from the William M. Bass Donated Skeletal Collection at the University of Tennessee Knoxville were used for this study. The individuals were born between 1929 and 1940, and died between 1987 and 2009. The range of age-at-death is 19 to 74 years with a mean of 46.7 years. Using the digitizer, measurements of the heights of the cranium, vertebral columns from C2 to L5 (TVH), the vertical space height between the anterior margin of the sacral promontory to the level that connects the superior margins of the left and the right acetabulae (AP-SMLRACE), articulated talus and calcaneus, and the physiological length of the tibia (Martin 2) were recorded. The physiological length of the femora was measured using an osteometric board because of the limited arm length of the digitizer. The interlandmark distances were calculated by Euclidian distances, in two or three dimensions, and trigonometry.

The use of five different locations on individual vertebrae (anterior, left, right, posterior, and the average of the left and the right sides), allows alternative means of calculating TVH in the event that individual vertebrae columns are incomplete. The spinal curve corrections were adapted from the Intermediate Index (DI) of Delmas as normal for all ages as well as under 42 years of age. Delmas' Dynamic Index (DDI) was used for individuals 43 years of age and older. These indices were used to evaluate additional curve reductions to determine if they should be applied to older ages. The CCST heights were obtained by subtracting the Total Skeletal Heights (TSHs) from the Adjusted Cadaver Stature (ACS)

The relationships among CCSTs, TSHs, and age were investigated using the Pearson correlation coefficients. The results showed the CCSTs had negative correlations ($r=-0.206$ to -0.409) to age whereas the TSHs had almost no correlation ($r=0.012$ to 0.1) to the CCSTs. Next, the test on residuals of Partial correlation was examined and confirmed that there are no influences after controlling on the TSHs. These results indicated that the individuals who have tall TSHs do not necessarily have thicker CCSTs. Therefore, the ten regression formulae for estimating CCST heights from all five locations, DI, and DDI were constructed from only age (e.g., Anterior CCST (mm) $= -0.853 \cdot \text{age} + 179.58$, 95% PI (Point Estimate $\pm 54.13 \sqrt{1.026 + ((\text{Age} - 46.74)^2 / 7107.69)$). ACS to Estimated Biological Statures (EBS = TSH + CCST) were tested by Paired *t*-tests on the ten models which ranged from -0.006mm to 0.0197mm with 95% lower CI between -8.269mm to -8.970mm and upper CI between 8.309mm to 8.947mm ($p>0.996$). Lastly, four independent samples of war casualties (European ancestry)

from Joint POW/MIA Accounting Command-Central Identification Laboratory (JPAC-CIL) were examined for accuracy and bias from antemortem stature records. The results showed Accuracy: 10.76 to 13.09mm, Bias: -0.71 to 17.13mm .

More accurate individual total skeletal heights TSHs were obtained using the digitizer, specifically when the S1 height was replaced with AP-SMLRACE and the physiological length of the tibia (Martin 2) from Fully was used. This methodology will be useful for skeletons that lack records of biological stature to create stature estimation equations from single and multiple elements in order to increase group-specific and generic stature estimates.

Standing Anatomical Position, CCST Components, Digitizer

H100 Examining Admixture and Secular Change in a Cranial Sample From Southern Japan

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After attending this presentation, attendees will better appreciate how cranial vault morphology has changed through time to result in the morphological complex observed in modern Japanese populations.

This presentation will impact the forensic science community by providing quantification of how the neurocranium can be used to understand population history in a specific East Asian sample, which can ultimately be used to elucidate how evolutionary processes such as gene flow and secular change can be better identified in populations that are often encountered in forensic contexts within the United States.

Results of this study will also be used to examine how discriminant function analyses using reference samples provided by FORDISC® 3.0 software can misclassify Hispanic individuals as representative of modern Japanese populations.¹

A multitude of research exists that investigates cranial variation of prehistoric and modern Japanese populations. Previous studies that employ multivariate analyses to two dimensional cranial measurements have addressed hypotheses regarding the population history of the entire Japanese archipelago, which has mainly focused on the biological distance and relationship of the prehistoric Jomon of Japan, Eneolithic Yayoi Agriculturalists, and modern Japanese groups within the context of the craniofacial form.^{2,3} While the seminal works of the last several decades have elucidated much of the population and migration history of Japan as a whole, less emphasis has been placed on the examination of skeletal samples representative of specific geographic areas and temporal continuity.

This study is one of the first to examine cranial variability utilizing the skeletal collection housed at Kyushu University in Fukuoka, Japan. This unique collection curates skeletal samples from Kyushu Island and nearby locales that are temporally representative of a nearly continuous sequence from the prehistoric Jomon culture to specimens from the 20th-century. Thus, using this temporally deep sample allows for the testing of several hypotheses that build upon recent findings that suggest the neurocranium correlates more significantly with neutral genetic traits and thus is a better predictor of population affinity.^{4,5}

This study employs multivariate analyses, including principal component analysis, discriminant function analysis, and procrustes superimposition, to analyze three-dimensional coordinate data and two-dimensional linear measurements to allow for a thorough examination of the cranium in which the skeletal

modules of the cranial base, neurocranium, and viscerocranium are examined congruently and separately. Eighty-five cranial landmarks were collected using skeletal samples spanning from the prehistoric Jomon and Eneolithic Yayoi agriculturalists to the Edo and recent Meiji periods, all excavated within Kyushu island or neighboring prefectures. Examination of the cranial base and neurocranium without the viscerocranium was employed to test the hypothesis that migration and gene flow events during the Yayoi period in southern Japan could be better identified than with an emphasis of facial dimensions described by previous studies. This study hypothesizes that an intermediate morphology resulting from gene flow from these distinct populations may be identifiable in the later skeletal series of the Kyushu samples when emphasizing shape and size variables of the cranial base and vault. These variables were also used to examine morphological trends evident over geographic time and space within the southern Japanese archipelago to build upon the secular change work carried out by previous studies.⁶

Results indicate that the removal of facial landmarks and concentration on dimensions associated with the vault and base suggests that skeletal samples subsequent to the Jomon and Yayoi periods exhibit cranial morphology intermediate to the parental samples. These findings lend support to the dual structure hypothesis proposed for the peopling of Japan, which posits that admixture rather than replacement occurred between the Neolithic Jomon and Eneolithic Yayoi cultures.³ Differing results are found with the inclusion of facial variables, which makes interpretation of relationship between temporal groups more difficult, as more variation is identified within groups when nasal and maxillary landmarks are included. These results support the hypothesis that the cranial vault may provide better resolution for population affinity as well as provide support for population continuity in the context of prehistoric southern Japan. These preliminary findings will be used to further elucidate how admixture between ancestral populations can impact cranial dimensions in forensically relevant samples, such as Hispanic populations, that are often encountered in the United States.

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Cranio-metric, Secular Change, Japan

H101 Sex Estimation of Juvenile Human Skulls Using 3D Geometric Morphometric Assessment of Craniofacial Architecture

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After attending this presentation, attendees will gain an understanding of using 3D geometric morphometrics to analyze shape differences in the human skull, as well as how these subtle differences can be used to investigate sexual dimorphism and to discriminate between male and female juvenile humans.

This presentation will impact the forensic science community by contributing to current forensic anthropological research regarding sex estimation from juvenile human skeletal remains, specifically by adding insight into particular morphological and growth trajectory differences between the sexes during development.

Although there is a coherent body of literature and consensus on the morphological presentation of sexual dimorphism in the adult human skeleton, there is still debate about the ontogenetic origins and proper methodology for assessing sexual dimorphism and sex differences in the juvenile humans. Previous studies have focused on measuring sexually dimorphic skeletal elements and features that are commonly used to estimate sex in adults, but because these features are sex characteristics that are neither present nor complete until adolescence at the earliest, these methods enjoy less success when applied to immature skeletal remains. This lack of consensus is compounded by findings that the greatest amount of sexual dimorphism in the skull occurs in areas that attain adult size late in ontogeny. Therefore, applying methods for sex estimation built on adults is problematic for use on juvenile humans, and estimating the sex of a juvenile individual from skeletal material remains tenuous at best.

Using Enlow's mammalian craniofacial architectural relationships, Bromage demonstrated that there are statistically significant morphological differences in female and male juvenile chimpanzee crania.¹ These differences were measured using 2D lateral cephalograms and centered on the spatial relationship between the cranial base and the facial skeleton. In the present study, Bromage's methodology is modified and expanded to collect 3D data from juvenile human Cone Beam Computed Tomography (CBCT) scans. These data are then analyzed using geometric morphometrics in order to investigate craniofacial sexual dimorphism during ontogeny.

A sample of CBCT scans derived from Australians 6-13 years of age were analyzed ($n=50$ males and $n=48$ females). Three-dimensional landmarks for 46 craniofacial architectural points were measured, including 12 midline landmarks and 17 bilateral landmarks, using Analyze 11.0 software were the locations of all 46 points for the entire sample independently identified. Points that could not be agreed upon within 1.6mm — or 4 voxels on the 0.4mm/voxel image — were discarded from analysis.

Principal components analysis, discriminant function analysis, and regressions revealed that the craniofacial architecture of male and female juvenile humans in this sample is very similar, though often not statistically significantly different; however, there are appreciable trends in variation between the sexes and in different age groups that warrant further examination.

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Geometric Morphometrics, Sexual Dimorphism, Juvenile Skull

H102 Validation of a Method of Sex Determination Based on Sterna From a Sample of Modern Colombian Mestizos

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The goal of this presentation is to demonstrate the applicability of a method of sex determination using discriminant function analysis developed in 2001 by Puerto based on osteometrics of the sternum of a modern Colombian sample.¹ The study presented tested the method using a new sample from a different region of the country to validate its use in widespread forensic contexts in Colombia.

This presentation will impact the forensic science community by demonstrating the necessity of testing sex determination methods based on osteometrics. The results of the formula applied do not support the use of this method in forensic practice.

The method developed by Puerto used a sample of 100 sterna (50 females and 50 males) from medicolegal cases from the National Institute of Legal Medicine in Colombia. All the individuals with age-at-death of 20 to 65-years-old were born in the Andean region of the country. Five measurements were obtained: manubrium length (X1); manubrium width (X2); body length (X3); combined length of manubrium and body (X4); and, width of body in 1st sternbrae (X7). Using multivariate analysis with the simultaneous equations method and the stepwise method, Puerto developed two main discriminant function equations for sex determination: (1) $Z = (0.119 \times X2) + (0.060 \times X7) + (0.064 \times X4) - 15.841$; and, (2) $Z = \{(0.0640 \times X2) + (0.040 \times X3) + (0.182 \times X1)\} - 15.360$.

In each of the previous equations, the sectioning point was 0, with females falling below 0 and males falling above 0. Results after applying the formulas to the sample were as follows: the first equation had a success rate of 90% and second equation had a success rate of 87%.

With the aim of testing the method developed by Puerto, this research used a sample of 50 sterna (25 females and 25 males) from a modern skeletal collection from the Prosecutor General's Office in Colombia. Age-at-death was 20 - 65 years old, and all individuals were born on the Caribbean coast of the country. Measurements were taken following Puerto and were considered as independent variables. Using the sex as the dependent binomial variable, an Analysis of Variance (ANOVA) test was conducted using the statistical software Statgraphic® 5.1. Sex of the individuals was estimated using the above discriminant functions, and subsequently real sex was compared to estimated sex. Results indicate that the first equation classified correctly 71% of the females and 46% of the males, which means a success rate of 52%. The second equation classified correctly 100% of the females and 46% of the males, which means a success rate of 50%. These results are lower than the ones presented by Puerto. This shows that these formulas are not dependable for identification purposes in Colombia.

This study indicates that population heterogeneity may be at play here, given the metric differences in the sterna of individuals from different parts of the country, although this hypothesis remains to be tested. Also, while this method needs to be subjected to further scrutiny, it presents the necessity to investigate the influence of factors such as migration and secular change among other factors, to understand how these are affecting the osteometric sex expression of Colombian sub-populations. This research expects to contribute to the standardization process of methods in Colombia, which ultimately will improve the quality of forensic analyses in a country that still has thousands of victims to be identified.

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Sex Determination, Sternum, Colombian Population Standards

H103 Application of Ischium-Pubis Index for Sex Determination Using 3D Models

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This goal of this presentation is to compare the accuracy of three methods to calculate ischium-pubis index. The new method using the deepest point of the acetabulum outperformed the traditional method of the point where the innominates meet.

This presentation will impact the forensic science community by presenting previous studies about ischium-pubis index that reported the method is susceptible to subjectivity, as the landmark in the acetabulum can be varied. As being able to mark the deepest point of the acetabulum through the 3D metric method, sex determination from ischium-pubis index has become an objective way of practicing forensic anthropology.

For sex estimation with the pelvis, ischium-pubis index is commonly chosen along the greater sciatic notch. This index is the percentage value of the pubic length divided by the ischium length and knowing that females have greater value than males. The pubic length is the distance from the upper edge of the pubic symphysis to the acetabulum, and the ischium length is the distance from the most distinct point on the ischial tuberosity to the acetabulum. The point in the acetabulum is customarily marked on the point where the three parts of the pubic bone meet; however, due to irregular shape of the acetabulum, there is a possibility of it being marked differently due to the observer. Thus, a landmark which we studied is more objectively marked and the ischium-pubis index is applicable to the Korean population.

This study focused on 103 sides (52 female and 51 male) of the Digital Korean Database stored in the Catholic Institute for Applied Anatomy. Landmarks chosen near the acetabulum to measure the pubis length and ischium length were: the point where the pubis, the ilium, and the ischium meet; the deepest point of the acetabulum; and a point on the acetabular border. Mimics® version 15 was used to enable three-dimensional measurement by a template programmed for this study. There were nine measurement items related to the distance and angle between each point. Discriminant analysis for sex determination was completed by SPSS version 20.0.

The method using the deepest point of the acetabulum showed the highest accuracy of sex estimation compared to the traditional method using the contacting point of the three innominates and another method of upper border of the acetabulum. Therefore, it is suggested through this study that utilising the deepest point of the acetabulum is suitable for calculating the ischium-pubis index. For this study, the mean ischium-pubis index of males was 98 and

of females was 114, and the demarking point of both sexes was 106.

Sex Determination, Ischium-Pubis Index, 3D Models

H104 A Comparison of the Kiales *et al.* (2012) and Phenice (1969) Methods of Sex Estimation on a Modern Colombian Sample

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After attending this presentation, attendees will be able to compare and contrast the sex estimation methods outlined in Kiales *et al.* with those of the Phenice method, as well as evaluate the application of these methods to a non-European American population sample.^{1,2} Attendees will be presented with a comparison of results obtained by the application of both methods on the Antioquia Modern Skeletal Reference Collection, a documented modern sample housed in Medellin, Colombia.

This presentation will impact the forensic science community by providing data on the application of a new method of sex estimation developed by Kiales *et al.* on a Colombian sample, and its efficacy in comparison to the widely used Phenice method.

Kiales *et al.* modifies the Phenice method by expanding the scoring scheme and incorporating statistical analysis to provide a probability that an individual is male or female. It is posited in this presentation that the Kiales *et al.* method might be more suitable for forensic cases where testimony in a court of law is necessary; however, it does not perform better than the Phenice method when applied to a modern Colombian sample.

Sex estimation is an important component of the biological profile during analysis of human skeletal remains. The Phenice method of sex estimation is based on the scoring of three distinct pelvic traits known as the ventral arc, subpubic concavity, and ischiopubic ramus, as feminine (1), masculine (3), or ambiguous (2), with the final determination of sex dependent on the average score obtained. This limited scoring system does not represent the variation exhibited by individual males and females, as most individuals do not exclusively exhibit the traits of one sex. Thus, sex estimation using the Phenice method is often reliant on professional opinion and experience. Kiales *et al.* expand the scoring system, allowing a possible five scores for each of the three pelvic traits. The scores of each pelvic trait are plugged into a logistic regression equation that produces a weighted score, which is then used to generate a probability. The sectioning point of the weighted scores is 0, with negative scores having a higher probability of being female and positive scores having a higher probability of being male. The two methods were tested on a sample of 50 individuals (39 male, 11 female) from the Antioquia Modern Skeletal Reference Collection curated in Medellin, Colombia. The sample age-at-death ranged from 17 to 99-years-old, with an average age of 49.9-years-old.

Results show a significant difference in overall accuracy rates for both methods (66% for Kiales *et al.*, 82% for Phenice). The Kiales *et al.* method resulted in a 45% accuracy rate for females and 72% accuracy for males. Using the Phenice method, accuracy rates were 78% for females and 83% for males. Three individuals were scored as ambiguous when using the Phenice method. In these cases, two out of three individuals were correctly assigned using the Kiales *et al.* method. In the instances where Kiales *et al.* provided incorrect sex estimation but Phenice was correct, a tendency for the Kiales *et al.* method to score male individuals as female was noted.

These results suggest that the Phenice method is preferable to the Kiales *et al.* method, though in cases where the pelvic scores are ambiguous, the Kiales *et al.* method proves useful. Both methods yielded higher accuracy rates for males; however, this may be attributed to the uneven sex distribution in the sample, with males far outnumbering females. More research is needed to determine the validity the Kiales *et al.* method and the degree to which it improves upon the Phenice guidelines. A larger and more evenly distributed sample would more accurately represent the applicability of the Kiales *et al.* method. Future testing in Colombia is necessary as more documented females become available.

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Sex Estimation, Forensic Anthropology, Colombia

H105 Sexual Dimorphism in the Cementoenamel Junction of American Blacks

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After attending this presentation, attendees will have a better understanding of the ability to estimate sex from Cementoenamel Junction (CEJ) diameters and the reliability of these measurements.

This presentation will impact the forensic science community by providing accuracy rates of CEJ measurements in sex classification and determining which measurements are most reliable for sex estimation methods.

Tooth enamel is composed of the hardest tissue in the human body, resulting in exceptional preservation in the various contexts of forensic cases. For this reason, tooth dimensions are a valuable tool in forensic analyses. While a number of studies have analyzed the utility of tooth crown dimensions in sex estimation methods, measurements at the CEJ have not been thoroughly investigated. This region of the tooth is particularly resilient, as the area is protected by alveolar bone. Additionally, the junction of the cementum and enamel is clearly delineated, providing a reliable location for measurements. A recent study has shown that CEJ measurements significantly vary between Japanese males and females, and also between Japanese females and South Asian individuals when estimating ancestry.¹ However, CEJ differences between modern American populations have not yet been investigated. The goal of this study is to determine if CEJ measurements can be utilized in sex estimation of American populations, and if so, whether maxillary or mandibular measurements are the most accurate in estimation. Finally, this study examines which singular tooth measurement is best suited for estimating sex.

Data were collected from 20 Black females and 21 Black males from the Hamann-Todd skeletal collection following Hillson's measurement definitions.² Measurements included the buccolingual and mesiodistal diameters of the maxillary and mandibular left canines, left third premolars, and left second molars. Linear Discriminant Function Analysis (DFA) in *FORDISC*® 3.1 was employed in this study and was used to maximize the mean differences between groups using Mahalanobis distances and the pooled within-group variance-covariance matrix.³

When all measurements were included in a discriminant

function analysis using a forward mean stepwise selection method and leave-one-out cross-validation method, the results indicate a 94.3% correct sex classification rate. The six measurements retained in the discriminant function, and hence the most sexually diagnostic, were the mesiodistal and buccolingual diameters of the mandibular canine, buccolingual diameter of the maxillary canine, the mesiodistal and buccolingual diameters of the mandibular third premolar, and the mesiodistal diameter of the maxillary third premolar. When the mandibular and maxillary CEJ dimensions were analyzed independently, the mandibular measurements performed better than the maxillary measurements (91.2% and 81.1% correct classification, respectively). Overall, the canine measurements were the most sexually diagnostic. In particular, the mesiodistal diameter of the mandibular canine displayed the greatest weight in all analyses. A univariate analysis using only this measurement resulted in an 85.7% classification rate.

As indicated by this study, tooth root dimensions at the CEJ can be successfully used in estimating the sex of an unknown individual. Combining various CEJ measurements in DFA results in a higher percentage of correctly classified individuals than using maxillary or mandibular measurements alone. This study indicates that CEJ measurement may provide a valuable resource in forensic sex estimation methods with accuracy rates comparable to methods utilizing the pelvis.

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Sex Estimation, Odontometrics, Cemento-enamel Junction

H106 Can We Use Dental Wear for Age Estimation of Modern Americans?

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After attending this presentation, attendees will understand that dental wear is a biased aging indicator in the modern American population; therefore, aging methods based on dental wear can hardly be applied in a forensic context.

This presentation will impact the forensic science community by providing empirical evidence through sophisticated statistical evaluation stressing the inability of dental wear being used as an accurate aging indicator in modern, contemporary Americans.

Contrary to the popularity of using dental wear as an aging indicator in archaeological materials, contemporary forensic communities have had very few publications regarding this subject. Furthermore, no research was found that specifically used the examination of dental wear as an independent aging indicator for modern populations. The goal of this study is to evaluate the accuracy versus inaccuracy of dental wear as an aging indicator in modern American populations and, therefore, provide forensic anthropologists with a theoretical basis for the use or disuse of dental wear when constructing biological profiles for human remains.

As a pilot study, the dentition of 73 modern White male

skeletons in William M. Bass donated collection at the University of Tennessee, Knoxville were examined. The age of the samples is normally distributed with the mean of 52.71 ($SD=13.03$) and the range of 25 to 78 (Kolmogorov-Smirnov test, $D(73)=.066$, $p>0.05$). Dental wear was assessed according to the eight criteria of Smith, with the exception of the third molars, and average scores were calculated for each tooth group (i.e., incisor, canine, premolar, and molar).¹ For statistical evaluation, a total of ten statistical analyses were performed: four simple regressions of the score of each tooth group on actual age; one multiple regression of the scores of four tooth groups on actual age; four logistic regressions of the score of each tooth group on age categories; and one logistic regression of the scores of four tooth groups on age categories. Age categories were defined following Hrdlička, by which samples were assigned into one of the four age groups of 26-34, 35-50, 51-64, and 64+ years.² The appropriateness of dental wear as an aging indicator was assessed based on the Pearson's correlation coefficient (r), R^2 and Nagelkerke's R^2_N .

In the results of simple and multiple regression analyses, the highest r and R^2 were 0.405 and 0.164, respectively, when all tooth groups were regressed on actual age. Since the adjusted R^2 in this model was as low as 0.06, it could be concluded that some unnecessary predictors exaggerated R^2 ; therefore, this regression model cannot be generalized. In the results of logistic regression analyses, the highest Nagelkerke's R^2_N was 0.25 which was obtained when only incisors were included in the model. Scatter plots revealed that this low relationship between dental wear and age was due to slight dental wearing in older samples rather than severe dental wearing in younger samples.

Obviously, any values of r , R^2 , and Nagelkerke's R^2_N in this study did not satisfy the standard ($r>0.9$ or $r>0.7$) to yield an accurate assessment recommended by Bocquet-Appel and Masset and Lovejoy *et al.*^{3,4} Moreover, considering that the r values of this study are lower than other studies regarding popular aging indicators such as pubic symphysis, auricular surface, and the 4th rib end, using dental wear as an independent aging indicator can hardly be justified.

Despite any possible limitations due to incorporating only White, male samples in this research, results of the statistical evaluation have revealed the inability to use dental wear as an accurate age indicator for modern contemporary Americans. Therefore, this research has a two-fold practical significance. It stresses that the use of skeletal features (i.e., dental wear) can be context-dependent (e.g., archaeological versus forensic context) and also provides empirical evidence to caution, if not discourage, forensic anthropologists when using dental wear to estimate the age of human remains.

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Dental Wear, Aging Indicator, Modern Americans

H107 Drug Use, Homeostasis, and the Estimation of Age-at-Death From Skeletal Remains

The goal of this presentation is to discuss degenerative skeletal aging as a function of biological homeostasis and the potential effect of disruptions to the normal aging process caused by drug and alcohol use.

This presentation will impact the forensic science community by demonstrating that no significant differences in biological age were found between individuals suspected of using drugs and alcohol and those without evidence for drug or alcohol usage.

Age-at-death is routinely estimated as part of the biological profile in order to facilitate the identification of an unknown decedent. The estimation of adult degenerative age is typically conducted using a few, relatively well-documented areas of the human skeleton. The most common are the pubic symphysis and sternal rib ends.¹ While there are a number of publications discussing age-related changes in the pubic symphyseal face and the sternal ends of the fourth rib, the underlying causes of these age-related processes are not well understood; however, neither is degenerative aging in general.² Much of the degenerative aging process occurs as a function of maintaining bodily homeostasis and disruptions in homeostasis can produce significant tissue pathophysiology.³ Disruptions caused by drug and alcohol abuse have been documented as resulting in pathological conditions such as osteosclerosis as a function of a stressed neuroendocrine system.^{4,5} However, while the potential skeletal effects of drug and alcohol usage have been documented in single case study contexts, this has yet to be demonstrated on a large population.

To examine the effects of drug and alcohol use on biological markers of age, a sample of 579 documented individuals, processed through Maricopa County Forensic Science Center (FSC) in Phoenix, Arizona, were examined.⁶ Data on the pubic symphysis and the sternal rib ends were collected for individuals with ages ranging from 18 to 99 years. Drug or alcohol use for individuals was established through the presence of drug- or alcohol-related items at the location the decedent was recovered from or from witness accounts. The available documentation was not detailed enough to establish chronic drug or alcohol abuse, but only to infer usage in general. To examine the potential effects of drug or alcohol usage on the degenerative aging process, correlations for each age indicator and combined indicators were examined. Based on the available individuals, samples were divided into drug/alcohol usage (n=94) and non-usage (n=483) and further sub-divided into Younger Adults (aged 18-39 years) and Older Adults (aged 40+ years).

While all age marker correlations decreased with advancing age and all standard errors increased with age, results indicate that drug/alcohol use in the present sample, as suggested via witness statements or the presence of drugs or alcohol, did not affect the ability to estimate age. Correlations were very similar between the two samples; however, the standard errors were comparatively low for drug/alcohol users aged 40+ years; this could be an effect of sample size. Further research using a similar approach on a sample with more detailed background information on drug and alcohol use is necessary and may be able to differentiate individuals suffering from chronic abuse which may significantly affect bodily homeostasis and aging.

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Drug Use, Homeostasis, Age-at-Death

H108 Biological Sex and Ancestry Uniqueness of Fingerprint Minutiae: An Anthropological Approach

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After attending this presentation, attendees will see the potential of focusing on the biological basis of the permanence and uniqueness of fingerprints for maintaining the objectivity inherent to fingerprint comparison.

This presentation will impact the forensic science community by demonstrating that fingerprint development is rooted in biology and, therefore, so is the use of fingerprints as a method of identification.

Dermatoglyphics have been studied extensively in physical anthropology to examine the heritability of friction skin traits and inter-population variation. Within the field of forensic science, dermatoglyphics of the fingers and palms have been used in comparisons to match unknown latent prints to complete prints of known individuals for over 100 years. Though studying the same patterns of friction skin, anthropology and forensic science have considered different variables and research questions. The majority of the previous anthropological studies have tested these population relationships on Level 1 Detail (e.g., pattern type, total ridge count), while forensic scientists focus on individual uniqueness of Level 2 and 3 Detail (e.g., minutiae and pores, respectively). Therefore, the results of anthropological studies are largely irrelevant to latent fingerprint analysis, where identifications are made based on comparisons of Level 2 and 3 Detail. Given the questions raised by the National Academy of Sciences Report of 2009 about the subjectivity of latent print examination, research efforts need to be made to focus on the objective aspect of fingerprints, namely their unique biological development. Using a physical anthropological perspective is one way to do this. The present study applies methodologies developed in physical anthropology for quantifying Level 1 fingerprint traits to Level 2 Detail, and tests whether population, sex, and/or pattern type has a significant effect on the number and type of minutiae. Five types of minutiae were analyzed, which include bifurcations, ending ridges, short ridges, dots, and enclosures, as well as a variable for the sum total of all minutiae. Each type of minutia was visually counted on the right index finger of a total 115 individuals (n = 29 African American ♀; n = 29 African American ♂; n = 29 European American ♀; n = 28 European American ♂). A Multivariate Analysis of Variance (MANOVA) was used to analyze the overall effect of sex, pattern type, and population on the minutiae variables. Results of the MANOVA show that only population significantly affects minutiae (p-value=0.019). In addition, contrast statements in the MANOVA model, as well as canonical correlation, were performed in order

to identify which of the minutiae variables are being influenced by the main effect of population and showed that ending ridges and dots are significantly influenced. Logistic regression was used to explore whether ending ridges and dots can be used to classify individuals into ancestry groups. Results show that neither of these minutiae can be used to predict population. The results of this study suggest that fingerprint development is driven by a complex biological system that is influenced by a wide variety of factors, both genetic and environmental. This finding partially explains fingerprint uniqueness and emphasizes the importance of including biology in the process and explanation of fingerprint comparisons.

Minutiae, Sex Variation, Ancestry Variation

H109 The Pig-of-the-Month Club: Seasonal Variation in Decomposition in a High-Altitude Desert

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After attending this presentation, attendees will have become familiar with aspects of decomposition seen in a high-altitude desert environment.

This presentation will impact the forensic science community by examining postmortem interval estimates in arid climates.

Variables thought to significantly affect decomposition, as measured by a total body score, are time, temperature, and access of the body to insects.¹ This project set out to test how well these suppositions applied to a high-desert environment in western Colorado and whether seasonality made a difference in decomposition rate with time and temperature normalized through the use of Accumulated Degree Days (ADD).

The project took place at Colorado Mesa University's Forensic Investigation Research Station which lies in Mesa County, Colorado, at an altitude of approximately 4,700'. In 2012, the National Weather Service recorded a total of 4.53 inches of rain a year in nearby Grand Junction, an average rH for the year of 42%, and 283 fair days, 62 partly cloudy days, and 21 cloudy days.² By any measure, this area is dry and sunny.

Beginning in October 2012, one deceased swine a month was deposited in a fenced outdoor facility. A HOBO® weather station within the facility measured temperature, precipitation, rH, solar radiation, wind speed, and wind gusts on an hourly basis. Decomposition was scored on the basis of gross morphology using the Total Body Score (TBS).³

Maggot masses were seen on all pigs, although at different times post-deposition, reflecting the annual change in temperature. Approximately 50% of the pigs ruptured during bloat. This is seldom seen in human casework in the region, and then generally as a result of injury rather than part of the normal decomposition process. An ongoing study of 150 cases of decomposed individuals from Mesa County from 2007 to 2012, currently being conducted shows no human cases with rupture. As the rupture allowed insects' additional access to the soft tissues inside the body, this may be a difference in decomposition between the humans and pigs worth further exploration.

Pigs varied in the time it took to reach a TBS of 24. Most pigs plateaued at a score of 24. At the score of 24, generally the carcass consisted of a leathery cover with no visible skeletal elements or insect activity. When the carcass was rolled over, active decomposition was seen underneath. In some cases, adipocere formed under the carcass. Pigs moved past the score

of 24, but not in their order of deposition, as would be thought if time, temperature, and access of the body to insects were the sole determinants of decomposition.

In addition to time and temperature, the rate of decomposition appears affected by the humidity and available moisture in the initial phases of decomposition. The faster the tissue desiccated past the point where fly larvae either no longer wished to, or could, consume the tissue, the slower the decomposition process. If the tissue stayed moist, it could be consumed by the fly larvae. As the tissue dried out, the mechanisms that appear responsible for the removal of the additional tissue were other species of insect larva and the gradual desiccation of the tissue to a very thin, paper-like consistency where it would break apart under minimal pressure.

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Taphonomy, Desiccation, Mummification

H110 CODIS and Anthropology: A Partnership for the Identification of Remains

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WITHDRAWN

H111 An Examination of the Relationship Between Asymmetrical Antemortem Tooth Loss and Asymmetrical Cranial Suture Closure

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After attending this presentation, attendees will be acquainted with the results of an examination of the possible relationship between asymmetrical antemortem tooth loss and asymmetrical cranial suture closure. In addition, attendees will understand the potential impact of asymmetrical cranial suture closure on the estimation of age-at-death.

This presentation will impact the forensic science community by providing a foundation for further examination of the causes of asymmetrical cranial suture closure, thus facilitating the refinement of suture-based age-at-death estimation methods.

When human remains of forensic significance are recovered in a fragmentary condition, cranial suture-based age estimation methods can be extremely useful in establishing or confirming an age estimate. If only one half of the skull is recovered, however, bilateral asymmetry in suture closure could lead to an erroneous age estimate and hinder identification of the individual.

The objective of this research was to determine whether there was a relationship between asymmetrical antemortem tooth loss and asymmetrical cranial suture closure in a modern human sample. One hundred-one crania from the historic Hamann-Todd Collection and the modern Bass Collection were examined in this study. Degree of suture asymmetry was measured at thirteen suture sites in each individual. The Variable Points of Dental Asymmetry (PDA) was designed and implemented to measure asymmetry in posterior tooth loss. It was hypothesized there would be a positive correlation between points of dental asymmetry and degree of suture asymmetry. The variables age-at-death, sex, ancestry, and skeletal collection of origin were also evaluated for influence on degree of suture asymmetry. A mixed model analysis of variance was conducted using IBM® SPSS Statistics Version 21.¹ All effects were evaluated for significance at the $p < 0.05$ level.

The main effect of PDA on degree of suture asymmetry was not significant ($p = 0.393$). The main effect of age-at-death on degree of suture asymmetry was also not significant ($p = 0.969$). However, the interaction effect between suture site and age-at-death was significant ($p = 0.003$). The main effect of sex on suture asymmetry was not significant ($p = 0.277$). The mean degree of suture asymmetry in those of African ancestry was higher than in those of European ancestry ($p = 0.05$). The difference in suture asymmetry between the Bass and Hamann-Todd subsamples approached significance ($p = 0.064$).

This study did not confirm a relationship between dental asymmetry and suture asymmetry based on the insignificant main effect of PDA on suture asymmetry. However, further research into the possible relationship between asymmetrical antemortem tooth loss and asymmetrical cranial suture closure is warranted. The variable PDA may not be truly representative of asymmetry in antemortem tooth loss. A variable that better captures the complexity of the biomechanics of mastication may demonstrate a relationship with suture asymmetry. A new variable representing asymmetry in antemortem tooth loss will be designed and incorporated into the next stage of research.

Although the main effect of age on suture asymmetry was not significant, the significant interaction effect between suture site and age indicates that there was a relationship between suture asymmetry and age at some sites or in some age categories, but not others. This interpretation is supported by the literature, which suggests that some suture sites are much more likely to achieve advanced closure with age than others, and therefore have more opportunity to achieve bilateral asymmetry.^{2,3}

The insignificant effect of sex on suture asymmetry indicates an absence of sexual dimorphism in this study. The borderline significant difference in mean degree of suture asymmetry between individuals of European and those of African ancestry indicates that suture asymmetry varies between ancestral groups. The main effect of skeletal collection of origin approached significance, but this may be a result of bias introduced by the differing distribution of the subsamples according to age-at-death, sex, and ancestry. Alternatively, it could be evidence of secular change in degree of suture asymmetry.

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Cranial Sutures, Age Estimation, Antemortem Tooth Loss

H112 Undocumented Border-Crosser Deaths Recorded by the Pima County Office of the Medical Examiner From 1990 to 2013: Leading Causes of Death and Demographic Characteristics of Decedents

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After attending this presentation, attendees will gain an understanding of how changes in border enforcement policies contributed to increased Unauthorized Border-Crosser (UBC) deaths in southern Arizona in the early 2000s — an increase that has not waned over the past several years despite a decrease in unauthorized migration flows in the area.

This presentation will impact the forensic science community by emphasizing the importance of systematically collecting valid and reliable data on estimated migrant deaths along the United States-Mexico border.

Between 1990 and 2013, the Pima County Office of the Medical Examiner (PCOME) has examined the remains of 2,338 UBCs—over 95% of which have been examined since 2000. The increase in migrant deaths in southern Arizona has coincided with increased border control efforts in the region during the mid-to-late 1990s. This is especially notable considering that recently passed Senate Bill 744 includes border enforcement measures that likely will directly contribute to even more migrant deaths along the United States-Mexico border.¹⁻³ Because many of these migrants are currently unidentified, since 2000 the PCOME has examined suspected UBCs and created a “cultural profile” among these decedents to distinguish foreign nationals in transit from US residents.^{4,5} Rather than deter would-be migrants, border enforcement efforts have rerouted migration flows into more remote areas along the border, including southern Arizona, and most recently, South Texas. This assertion appears to be supported by data collected by the PCOME. Over 45% of confirmed UBC deaths have been due to exposure or probable exposure, followed by undetermined cause of death (36%), motor vehicle accident (9%), other miscellaneous causes (6%), and homicide (4%).⁶ A multivariate analysis using these data also confirms that specific causes of death tend to vary according to UBCs demographic characteristics. Another focus of the presentation will be to familiarize people with the demographic profile of UBCs and discuss how this profile has changed over time. Generally speaking, the typical UBC examined by the PCOME can be described as a male near the age of 30 from central or southern Mexico who died of exposure while attempting to avoid detection by U.S. authorities. However, there have been important changes in the UBC profile over the past several years. Today, UBCs are slightly less likely to be female, slightly older, and more likely to be from Central America than a decade ago. For instance, between 2006 and 2013 approximately 17% of UBC decedents were from a country other than Mexico, compared to only 9% between 2000 and 2005. Counts of UBCs examined by PCOME during 2013 to date are on pace to meet or exceed counts over the past several fiscal years. It is probable that the border enforcement surge outlined in recent legislation will increase UBC deaths along the southern

United States border, especially as the demand for immigrant labor increases with the recovery of the United States economy. This, coupled with what appears to be a changing demographic profile of UBCs, will undoubtedly pose many new challenges for jurisdictions along the border that historically have not had to contend with the issue of migrant deaths. It is important for professionals in jurisdictions experiencing an increase in caseloads of UBCs to have a working knowledge of the factors that contribute to unauthorized migration flow.

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Border Enforcement, Migrant Biological Profiles, Causes of Death

H113 The Missing Migrant Project: Forensic and Cultural Anthropological Expertise Combined

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After attending this presentation, attendees will have an understanding of the complex set of factors which lead to the lack of missing persons data in the United States-Mexico border context, and how the Missing Migrant Project can provide assistance with such investigations. In addition, attendees will gain a deeper appreciation of how forensic anthropologists and cultural anthropologists can collaborate productively in contexts where there are high numbers of missing and unidentified individuals.

This presentation will impact the forensic science community by introducing the Missing Migrant Project (MMP), which assists forensic anthropologists and others in the identification of suspected Mexican or Central American migrants dying within United States jurisdictions.

The MMP was founded in 2006 at the Pima County Office of the Medical Examiner (PCOME) to support that office in its investigation of the deaths of hundreds of border crossers each year. There are specific challenges to human identification in this context, both on the physical side and on the social side. Found

in remote desert areas, often highly decomposed or skeletonized, and often without identification media, deceased Undocumented Border Crossers (UBCs) are physically difficult to identify. On the social side, there are hundreds of families in search of the missing from over five countries who are limited in their search by issues relating to national jurisdiction, poverty, and fear. Although hundreds of UBCs are dying on United States soil, complex factors at play in this massive transnational migration northward mark the border forensic context as distinct from everyday medicolegal practice in the United States.

The MMP was created to collect, manage, and share missing migrant reports among medicolegal agencies, Non-Governmental Organizations (NGOs), and foreign consulates, and then to compare such reports to biological profiles of unidentified remains recovered in the desert areas of southern Arizona. Undertaken collaboratively between a forensic and a cultural anthropologist, the MMP approaches the UBC crisis anthropologically, leveraging a broader set of tools in a setting where many social factors directly affect the medicolegal context. Such collaborations are common in international forensic investigations, but are rare in the domestic setting. The success of the MMP in communicating with both families and forensic scientists, facilitating dozens of identifications, provides strong evidence that collaborations of this kind are useful in the domestic medicolegal context.

The MMP database currently contains records for over 1,500 missing migrants, with an increasing number last seen alive in border states other than Arizona. In 2012, the MMP and the PCOME were granted permission to enter missing person reports for foreign nationals into National Missing and Unidentified Persons System (NamUs), allowing automatic comparison against unidentified remains. The MMP has established working relationships with law enforcement agencies, foreign consulates, and humanitarian groups along the border and is growing with increased funding, staff, and resources. The MMP will take missing migrant reports from any agency, organization, or individual STET will engage with the family and others to collect the highest quality of information, and then enter all data into NamUs.

Forensic Anthropology, Cultural Anthropology, Undocumented Migrants

H114 Ancestry Estimation in Forensic Anthropology: Geometric Morphometric Derived Data vs. Traditional Craniometric Data With Special Reference to Hispanic Crania

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After attending this presentation, attendees will learn about geometric morphometric derived data as compared to traditional cranial measurements with regard to ancestry estimation.

This presentation will impact the forensic science community by suggesting changes in standard forensic anthropology data collection protocol for improved ancestry estimation.

The term Hispanic has been used frequently in forensic anthropology literature.¹⁻³ The term is ambiguous when considering the genetic variation of the different national origin groups (e.g., Puerto Rico, Mexico) that comprise this designation.^{4,5} Currently, little is known about the morphological variation of groups considered Hispanic due to lack of skeletal reference data. Previous research suggests that reference data with the designation Hispanic provides ambiguous classifications for estimates of ancestry using traditional cranial measurements or Inter-Landmark Distances

(ILDs).⁶ The ambiguous classifications are likely due to the non-population-specific nature of the data. Geometric Morphometric (GM) methods in forensic anthropology have received attention in recent years concerning the potential to improve ancestry estimations due to the focus on shape independent of size.^{7,8} The purpose of this presentation is two-fold; to test whether GM derived shape and size variables perform better than cranial ILDs for the purpose of ancestry estimation, using population-specific data from several Hispanic groups from Mexico and Guatemala.

Due to large-scale data collection efforts at the Pima County Office of the Medical Examiner in Tucson, Arizona, and two universities in Mexico, craniometric data from positively identified Mexicans are now available. Mexicans currently comprise over 65% of the United States Hispanic population with over 35% foreign born.⁹ Therefore, data used in the present analyses can be considered population-specific for the majority of Hispanics in the United States. The following groups were used in all analyses: Mexican (n=95); Guatemalan (n=71); American Black (n=61); and American White (n=247). Although Guatemalans represent a smaller percentage of the U.S. Hispanic population, the increase in migrant deaths along the United States-Mexico border from areas other than Mexico, especially in Texas, make it important to distinguish among various national origin groups.¹⁰

Due to small sample size, females were excluded from the current analysis. In order to test whether GM-derived variables perform better than ILDs, several Discriminant Function Analyses (DFAs) were run. Standard ILDs include those outlined in Jantz and Moore-Jansen, and additional ILDs outlined in Howells were also included.^{11,12} All ILDs were imported into FORDISC[®] 3.1 using the custom import function and two DFAs were run, one with standard ILDs and one with Howells' ILDs, both using the stepwise variable selection procedure.¹³ For the GM analyses, a total of 35 landmarks were selected to represent overall craniofacial morphology and to maximize sample size. Using MorphoJ, a Procrustes analysis was performed and principal components and centroid size were obtained.¹⁴ The first ten principal components and centroid size were run in a DFA using SAS[®] 9.3 to obtain cross-validated classification rates for comparison with rates obtained from ILDs.¹⁵

Cross-validated classification rates using standard ILDs are 61% for Mexicans, 75% for Guatemalans, 80% for American Blacks, and 89% for American Whites. When Howells ILDs were used instead of the standard ILDs, classifications increased with 76% for Mexicans, 85% for Guatemalans, 91% for American Blacks, and 87% for American Whites. Using GM-derived principal components and centroid size, classifications are 61% for Mexicans, 58% for Guatemalans, 60% for American Blacks, and 84% for American Whites. The highest classification rates are those obtained using Howells' ILDs rather than those considered standard. Further, classification derived from GM methods are the lowest. The Howells ILDs used in the DFA include multiple angles involving basion, bregma, prosthion, and dacryon, and fractions and subtenses along the frontal bone.

These results suggest: (1) ILDs outperform GM derived data for the purposes of ancestry classification; (2) Howells ILDs outperform the currently used standard ILDs, especially for distinguishing Mexicans and Guatemalans from each other and other groups; and, (3) Mexican's and Guatemalans differ in cranial morphology, suggesting that it is possible to distinguish among national origin groups considered Hispanic. These results further suggest that it is time to revise our current, standard data-collection protocol in order to keep momentum with our constantly changing U.S. population demographics.

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Forensic Anthropology, Ancestry Estimation, Hispanic

H115 Migrant Deaths: Complexity of Assessing Place of Origin

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After attending this presentation, attendees will better understand issues surrounding the identification of undocumented migrants.

This presentation will impact the forensic science community by presenting the utility of using geometric morphometrics to assess place of origin.

Since the start of the United States border enforcement

strategy "Operation Hold the Line" in El Paso in 1994, border movement patterns and points of entry at the U.S.-Mexico border have significantly changed from unauthorized entries into residential areas along the border to more inhospitable areas of the American Southwest.^{1,2} According to the U.S. Customs and Border Protection, as the U.S.-Mexico border-enforcement strategies continue to militarize, the number of migrants apprehended has decreased, but the number of deaths has remained stable (~368 per year), increasing the ratio of deaths to apprehensions to approximately 11 percent in 2011. The Pima County (Arizona) Medical Examiner (PCOME) investigated over 1,000 deaths between 2001-2007 resulting from unauthorized entry at the Arizona-Mexico border.³ In addition, the *Arizona OpenGIS for Deceased Migrants* application lists 408 migrant deaths due to all causes for the years 2010-2012.⁴

The 2010 U.S. Census reported that Hispanics/Latinos make up 8.4 percent of the population of North Carolina, 30.2% of the population of Arizona and 16% of the population nationwide. The countries/region-of-origin for North Carolina Hispanics/Latinos follow national trends with 60.9% from Mexico, 9% from Puerto Rico, 2.2% from Cuba, 2.0% from the Dominican Republic, 13% from Central America, 5.7% from South America, and 7.1% listed as "other." A better understanding of the region-of-origin of undocumented migrants could assist in the identification efforts that have become a major human rights issue in the U.S.

The purpose of this study is to: (1) explore the possible origins of Undocumented Border Crossers (UBC's) who died in transit investigated by PCOME and compare these data to unidentified decedents from North Carolina and Georgia whose distance from the border makes their migration status uncertain; and, (2) explore if the UBC's from Arizona differ morphologically from the North Carolina UIDs. The sample totals 356 individuals (UBC n=129; PCOME known n=28; North Carolina/Georgia UID n=22; Panama n=24; Peru n=7; Guatemala n=77; Cuba n=19, Mexico Merida n=44; Mexico Morton Collection n=6). Fourteen standard craniofacial homologous coordinate landmarks were used in the analysis. The shareware program MorphoJ developed by Klingenberg was used to translate, rotate, and scale all coordinates into a common coordinate system using a Generalized Procrustes Analysis or GPA. Group comparisons were explored with a Canonical Variates Analysis (CVA) of the newly derived Procrustes coordinates. Approximately 83 percent of the total variation is accounted for on the first four canonical variates. A Mahalanobis distance was used to examine the similarity between the groups and results show that all groups are significantly different from one another based on 1,000 permutations (p -values range from 0.01-<0.0001). Interestingly, the Arizona UBC are closest to the Guatemalan sample ($D=1.755$) followed by North Carolina/Georgia to Panama ($D=2.283$) to Arizona UBC ($D=2.352$) to Guatemala ($D=2.593$) and the Arizona PCOME sample is closest to the Arizona UBC ($D=3.219$). These results show that the 2010 Census may not adequately reflect the country of origin of undocumented migrants in the United States.

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Migrant Deaths, Place of Origin, Variation

H116 Utilizing Open GIS Software to Map the Deaths of Undocumented Border Crossers

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After attending this presentation, attendees will better understand the utility of Open Geographic Information Systems (OGIS) in the field of forensic anthropology for mapping the recovery location of many individuals over a large geographic area.

This presentation will impact the forensic science community by highlighting the advantages of GIS mapping software when dealing with a large number of human remains, many of them skeletal and incomplete, over a long period of time.

The Pima County Office of the Medical Examiner (PCOME) investigates all unattended and suspicious deaths, among others, for 12 of the 15 counties in Arizona. Of those 12, five counties are located either along the U.S.-Mexico border or directly north of them and are known to have active migrant routes. In total, these five counties cover over 27,000 square miles, most of which are sparsely populated and contain harsh desert and mountainous terrains. Since 2001, over 2,100 undocumented border crossers have perished in the Sonoran desert within the PCOME's jurisdiction. It is the goal of the PCOME to identify these individuals and eventually make possible the return of their remains to their family. Over the past decade, the identification rates at the PCOME have averaged 64% (or approximately 1,300 individuals) with most of these identifications primarily being made through facial recognition or fingerprint comparison.¹ The majority of the nearly 800 unidentified remains cases are either skeletonized or significantly decomposed, so that other techniques, such as DNA comparisons, must be used for identification. Oftentimes, these types of cases are not complete bodies or skeletons either due to animal scavengers consuming and scattering the remains or due to insufficient recovery techniques.

To fully comprehend the magnitude of deaths occurring in the Sonoran desert, the PCOME partnered with Humane Borders, Inc., a local Non-Governmental Organization (NGO), to develop a free, open, source, online mapping system to "grant access to high-quality downloadable spatial data regarding migrant deaths."² The end result, which can be accessed at www.humaneborders.info, provides three mapping tools to plot the deaths of undocumented border crossers in southern Arizona. All three are available to the public in both English and Spanish. The first mapping tool allows for either a case-specific search by a decedent's name, or more generalized searches using such criteria as year of death, manner of death, decedent sex, or a combination of the available parameters. The second provides a spatial bounding-box to search for a known decedent or group of individuals within a specific geographic area as defined by the user. The third mapping tool is structured so that an individual case can be mapped using the PCOME case number to determine its location relative to landmarks or other cases in the area. It is this last tool that is most useful to the PCOME in the search for other potentially related cases within the vicinity of any particular set of remains.

Before the creation of OGIS, remains found in different locations or at different times could only be re-associated through blind genetic profile matches performed periodically within the DNA laboratories' internal database. However, a search conducted using the OGIS platform can identify other cases for direct genetic comparison, so that the remains may be re-associated in weeks or months rather than years, and ideally before the remains are released for cremation or returned to the family. Since the launch

of the software in May 2013, two cases have been determined to be associated by PCOME Forensic Anthropologists, and DNA testing is in progress to prove if they are indeed from the same individual. As more cases become available on the OGIS website, many additional sets of remains are expected to be tentatively re-associated before being sent for DNA testing.

In conclusion, the use of geographic technology within the field of forensic anthropology opens up a new pathway for dealing with such disasters as that seen in southern Arizona. In addition, these resources are useful for other jurisdictions experiencing an increase in migrant deaths; not only for those investigating the deaths, but also for the families of the missing migrants searching for information.

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Undocumented Border Crossers, Migrant Deaths, Open GIS

H117 Skeletal Indicators of Stress: A Component of the Biocultural Profile of Undocumented Migrants

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The goal of this presentation is to inform attendees of consistent differences in levels of physiological stress between Undocumented Border Crossers (UBCs) and individuals who grew up in the United States. Attendees will be encouraged to apply those differences in their forensic casework as criteria to aid in classifying unidentified human remains as undocumented migrants.

This presentation will impact the forensic science community by adding to an established suite of biological features commonly observable in the skeletal remains of foreign nationals from Mexico and Central America.¹ The ability to distinguish undocumented migrants from American-born individuals is growing in importance as the number of border deaths continues to rise, and as migration patterns and the geographic distribution of border deaths shift and expand. Forensic anthropologists working in border jurisdictions and, increasingly, across the U.S. will more frequently find it necessary to differentiate between undocumented migrants and U.S. citizens.

Previous research at the Pima County Office of the Medical Examiner (PCOME) has demonstrated that, when compared to Hispanics born in the United States, foreign national Hispanics have elevated prevalence rates of dental caries and antemortem tooth loss, in addition to shorter stature.² The present research builds upon those results and expands the framework of analysis. Because individuals crossing the border often come from depressed socioeconomic circumstances and are disproportionately likely to have experienced nutritional deficiencies and infections during growth and development, this study hypothesized that skeletal indicators of physiological stress would appear more frequently and with greater severity in the remains of UBCs than in those of individuals born in the United States.

To investigate this hypothesis, skeletal samples from the PCOME and the Maxwell Museum of Anthropology were macroscopically examined for the presence of non-specific indicators of physiological stress that included porotic hyperostosis, cribra orbitalia, and linear enamel hypoplasias. The UBC sample from the PCOME comprised 130 adolescents and adults who died

while crossing the U.S.-Mexico border into Arizona. Six additional individuals analyzed at the PCOME and included in this study were identified as non-border crossers. The skeletons from the Maxwell Museum, which formed the majority of the samples of non-border crossers, included 45 adolescents and adults from documented forensic cases. Each individual was scored for the presence or absence and the degree of expression of the aforementioned stress indicators. Prevalence rates were compared between the UBC sample and the non-UBC sample using Pearson's chi-square tests or, when cell sizes fell below five, Fisher's exact tests. All calculations were made using SPSS 20.0.

Undocumented border crossers were found to exhibit greater prevalence rates of all indicators of physiological stress assessed in this research. A striking proportion (83/107; 77.6%) of UBCs exhibited porotic hyperostosis, although expression was minor in most cases. The difference in the prevalence of porotic hyperostosis between UBCs and non-UBCs (17.6%) is statistically significant ($\chi^2 = 50.988, p=0.000$). Additionally, the prevalence rate of linear enamel hypoplasias in UBCs (31.3%) was significantly greater than that in non-UBCs (11.1%) ($\chi^2 = 6.497, p=0.011$). While the difference in the prevalence of cribra orbitalia between the UBC sample (11.0%) and the non-UBC sample (2.0%) did not reach the $p<0.05$ level of significance ($p=0.070$), the results for this condition appear to reflect a meaningful pattern that is likely to be confirmed in the future as data collection continues and sample sizes increase.

Because porotic hyperostosis, cribra orbitalia, and linear enamel hypoplasias appear with greater frequency among undocumented border crossers than among U.S.-born individuals, their presence in skeletal remains recovered from modern, forensic contexts is noteworthy. Given that most migrants reach their destination within the United States, forensic anthropologists — regardless of whether or not they work in border jurisdictions — should take this suite of skeletal health indicators into consideration when contextual and/or sociocultural evidence suggests that an unidentified human remains case may represent an undocumented migrant.

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Skeletal Stress, Undocumented Migrants, Biocultural Profile

H118 The U.S.-Mexico-Central America Border Project: The Search for Missing Migrants

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After attending this presentation, attendees will understand factors leading to increased migrant deaths since 2000, obstacles to reaching identifications in these cases, and approaches used to improve exchange of information as well as possibilities for identifications.

This presentation will impact the forensic science

community by discussing collaborative cross-national forensic practice, with the potential to significantly reshape forensic practice in some places regarding unidentified remains thought to correspond to missing migrants.

The Argentine Forensic Anthropology Team's (EAAF's) Border Project, begun at the end of 2009, plans to create a Regional Committee on Missing Persons and Unidentified Remains to significantly improve the identifications of migrants among unidentified remains in the region. The project affects the southern United States border, Central America, and Mexico. This multiyear project responds to: the growing number of unidentified remains thought to correspond to migrants in the region; the need to improve in some cases the quality and number of identifications of remains; the reliance in many instances on an *ad hoc* case-by-case approach by forensic personnel, law enforcement, and consulates to reach identifications as opposed to a regional, coordinated forensic system to exchange information on a massive scale; and the frustrations sometimes experienced by both Mexican and Central American families searching for missing migrants. To this end, EAAF is pursuing two parallel programs.

The first is a pilot project for the creation of forensic data banks on missing migrants in migrant communities of origin, containing case files with all available information on missing migrants, including antemortem and background information for each case and genetic profiles from relatives. Data banks have governmental and non-governmental partners working together and have begun to develop as autonomous and sustainable institutions, collaborating at a regional level. So far, data banks have been created in El Salvador, Chiapas, Honduras, and an initial mechanism in Guatemala. Select cases have been included from Costa Rica and other Mexican states. In total, these data banks include nearly 500 cases of missing migrants, as well as almost 1,400 genetic profiles from relatives. Case information is compared against unidentified remains in U.S. and Mexican morgues that may correspond to missing migrants for identification purposes. Working with consulates and local morgues, EAAF has determined the identification of 43 migrants at this point in time, twenty-eight from El Salvador, nine from Guatemala, three from Honduras, two from Chiapas, and one from Costa Rica. Twenty-two of the remains were recovered in Texas, seventeen in Arizona, three in Mexico and one in California.

The second program consists of a strategic planning project to improve regional governments' responses and public policies regarding cases of missing migrants, working toward the founding of the Regional Committee on Missing Persons and Unidentified Remains. The strategic planning project involves: meetings among Mexico-United States border forensic institutions, state prosecutors' offices, Non-Governmental Organizations (NGOs), and other relevant institutions to discuss accessing and crossing of information on unidentified remains and missing migrants, the treatment of families searching for missing migrants and, where needed, improving local standard operating procedures for investigating cases of missing persons and unidentified remains; presenting a regional approach at relevant conferences and hearings, such as those arranged by the Inter-American Commission on Human Rights; and, finally, presenting information before intergovernmental bodies like the Organization of American Studies (OAS) and United Nations (UN) on the challenges and obstacles posed by inadequate governmental responses.

To date, the Border Project has contributed to reaching new identifications that would not have previously been made, many involving cold cases, as well as the use of scientific evidence in identifications that may previously have relied solely on circumstantial evidence. In addition, evidence collected from identifications are contributing in some cases to processes of accountability and justice for families of migrants, and to reform government policy regarding forensic involvement in the

identification of migrants. Significant challenges remaining for the Border Project include reaching agreements with further national, state, and local agencies to access information on unidentified remains, and in the context of the data banks, maintaining project continuity during governmental administrative transitions. EAAF's long-term goal is to assist in creating the Regional Committee on Missing Persons and Unidentified Remains, which will help coordinate the work of the forensic data banks for missing migrants, oversee an ongoing and systematic forensic comparison of all cases across international borders, improve regional exchange-of-data, and disseminate forensic best practices.

Migrants, Identification, Regional Collaboration

H119 Same Border, Different People: Development and Assessment of Sex Estimation Methods for Multiple Hispanic Populations

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After attending this presentation, attendees will be familiar with different methods used to estimate the sex of skeletal remains from Latin America along the United States-Mexico border. These methods are typically based on remains analyzed at the Pima County Office of the Medical Examiner (PCOME) in Tucson, Arizona, which consists largely of Mexican individuals.

This presentation will impact the forensic science community by proposing new techniques for sex estimation and assessing the effectiveness across Latin American populations.

U.S.-Mexico border deaths continue to occur at an alarming rate, and PCOME casework, both identified and unidentified, have served as the foundational sampling source for many methods for estimating the biological profile when the case is presumed Hispanic. While the PCOME sample and methods based on it are an invaluable contribution to the field, a major presumption when working with this sample needs to be tested: Does the PCOME sample adequately represent skeletal variation of populations encompassed within the grouping of "Hispanic?"

The PCOME case demographic is primarily Mexican nationals, thus methods based on this sample primarily represent Mexican population variation. However, deceased migrants have been identified as citizens of other Latin American countries as well, such as Guatemala and El Salvador. Based on a 22-year study of PCOME casework, data indicate that non-Mexican migrants experience a greater risk of death while attempting to cross the border, and as these individuals increasingly attempt the journey, there is even more of an importance to have relevant methods for the entire migrant population, not just Mexican migrants.¹ Because the deceased are mostly unidentified when the forensic anthropological analysis is performed, the region/country of origin is typically unknown or presumptive, and, due to population variation, using PCOME sample-based methods may yield a significant error when applied to non-Mexican skeletal remains. To test the assumption that methods generated from the PCOME sample can be successfully applied to other increasingly common migrant populations, this study applied a sex estimation method previously established by Tise *et al.* of postcranial sectioning points to a modern (1980s) Guatemalan Maya sample.² The results indicate a substantial decrease in accuracy for estimating sex correctly for the Guatemalan sample (males: 50-69% accuracy; six sectioning points) as compared to the accuracy reported by Tise

et al. when tested on an additional PCOME sample (75-86% for same six variables).² This disparity in accuracy suggests that inter-population postcranial variation is significant.

One approach to ameliorate this issue is to combine data from various Hispanic populations and generate new methods based on the expanded dataset, consisting of skeletal components that are both sexually dimorphic and display limited inter-population variation. While the classification accuracy will likely decrease in comparison to those methods based on single populations, it will be a more conservative approach when the individual is unidentified beyond a presumption of Latin American origins. Fowler and Perez developed a discriminant function for predicting sex of Guatemalan Maya (GUA) using four dental metric variables and found high accuracy.³ To test for inter-population variation, the Guatemalan dental data from this study were compared to dental metric data from PCOME. Significant differences in mean dental measurements between the two samples were present in 1 out of 13 measurements for males and in 0 out of 5 measurements for females (number of variables tested dependent on sample sizes). Inter-population similarities for dental metric data make it potentially useful for estimating sex when region of origin is unknown beyond Latin America. To test its utility, a linear discriminant analysis (4 dentition metric variables) was performed on the combined GUA and PCOME sample to infer its utility in estimating sex across multiple populations. For the initial sample, jackknifed classification success is greater than 77% for both sexes. To assess whether those misclassified mimicked the postcranial method's trend of incorrectly classifying GUA males as females, this study examined the population origins of misclassified males. Approximately two-thirds of misclassified males were GUA, while misclassified females were more evenly distributed between the PCOME and GUA samples. By developing and assessing the utility of postcranial and dental metric methods for sex estimation of Hispanics, this study answers the call for critically assessing methods to applied border-crosser casework.

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Hispanic, Sex Estimation, Dentition

H120 Craniometric Variation and the Challenges of Identifying Diverse Individuals in Florida

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After attending this presentation, attendees will be familiar with the craniometric variation among four diverse Caribbean and Latin American populations in relation to American White and Black

individuals in order to understand the biological distances based on cranial morphology. The results may assist in the understanding of the similarities and differences among the crania of these populations to incorporate into forensic anthropological casework.

This presentation will impact the forensic science community by offering research into human cranial variation among diverse populations to improve identification methods for these populations within the United States and, specifically, in Florida.

The state of Florida is commonly ignored as a border state when it comes to issues and challenges in migrant death investigations. However, undocumented workers frequently arrive in Florida for labor, such as on produce farms. Individuals considered Latino or "Hispanic" encompass the largest minority group within the United States. In the 2010 U.S. Census, the total number of individuals who identified as "Hispanic" was almost 309 million individuals, with almost 19 million of those individuals living in Florida, making Florida hold the third largest population, following California and Texas, which are both located along the U.S.-Mexico border.¹

This paper explores the craniometric variation among populations commonly found in Florida, including individuals from Mexico (n=134), Puerto Rico (n=78), Cuba (n=95), and Guatemala (n=87). Additionally, American White (n=115) and American Black (n=27) individuals were included from the Forensic Anthropology Databank, typically representing individuals who lived in the southeastern United States.

Eighteen standard cranial measurements were selected in a stepwise selection procedure and used for multivariate statistical analyses. Shape variables were created in SAS® 9.1.3 to include both males and females together allowing for an increase in total sample size. Mahalanobis D² and canonical discriminant analyses were performed to compare significant differences between the six populations. The results of the distance relationship suggests that the samples from Mexico and Guatemala together show the greatest biological distance from all other populations, primarily from Puerto Rico and Cuba, which supports regional differences among Latin America and the Caribbean. This distance is illustrated in a canonical variates analysis, illustrating 76.0% of the variation, showing that Mexico and Guatemala are separated on CAN1 (59.0% of the variation) from all other populations with a smaller maximum cranial length and greater bizygomatic breadth. While the samples from Puerto Rico and Cuba show less distance from the American White and Black populations, the Mahalanobis D² results and CAN2 (17.0% of the variation) demonstrate that Cuba is closest to the American Black population, with a greater basion-bregma height, and Puerto Rico is closest to the American White population with a shorter basion-bregma height.

Population histories through migration patterns and gene flow are factors that have contributed to biological variation throughout the history of Latin America and the Caribbean. This research explores the variation among diverse groups that are living in Florida and throughout the United States. By improving our understanding of the historical and current biological relationships among diverse groups living in close proximity, forensic anthropologists can better utilize ancestry data into their biological profiles for human identification.

Reference:

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Cranial Variation, Border Deaths, Identification

H121 Identification of Deceased Undocumented Immigrants Along the Texas Border

After attending this presentation, attendees will understand the increasing crisis regarding border crosser fatalities in the state of Texas as well as the complexities and current efforts in the identification of these undocumented immigrants.

This presentation will impact the forensic science community by documenting forensic science lacuna in the Texas border region and the achievements in building a border consortium of forensic scientists to ensure unidentified remains are properly reported, analyzed, and included in state and national databases. The purpose of this presentation is to provide as an example of the findings regarding the circumstances and disposition of 98 cases of unidentified immigrants recovered from Brooks County, Texas, and the coalition of scientists currently analyzing these cases.

There are an estimated 11 million undocumented immigrants currently living in the United States. The majority of these individuals entered along the U.S.-Mexico border. The United States has strengthened enforcement of immigration laws by adding personnel and technology in areas of high activity. The "prevention through deterrence" initiative aimed to increase the possibility of apprehension for illegal entrants enough to create an environment of unacceptable risk. This has resulted in a 78% drop in the total number of apprehensions since 2000. In addition, it has moved the major areas of illegal immigration from urban centers to remote areas of inhospitable terrain. Unfortunately, despite the reduction in apprehensions, the number of border deaths is increasing due to this change in migration routes. In 2000 there were 2.3 deaths per 10,000 people who were apprehended crossing the border that has increased to 16 deaths per 10,000 in 2012. This represents an almost 600% increase and should clearly be considered a humanitarian crisis.

In Texas, undocumented immigrant fatalities increased 127% in the Rio Grande Border Sector between 2011 and 2012. The majority of these cases occurred in Brooks County where there were 64 cases in 2011 and 130 cases in 2012. Brooks County is 944 square miles and has roughly 7,200 residents so it is a vast area with a small population. The county predominately consists of very large ranches. The Sheriff's Department has five officers and there are four Justices of the Peace (JPs) that attend to the deaths in the county. One hundred and thirty immigrant deaths last year are equivalent to a prolonged mass disaster for this county. Brooks County does not have a medical examiner or any resident forensic scientists. Given the small number of officers and JPs and no forensic personnel, most of these cases have not had autopsies, DNA sampling, or been included in missing person databases. When a body is not identified, the individual is buried in the Falfurrias Sacred Heart Burial Park. Last fall, Baylor University's Reuniting Families Project (RFP) began discussions with Brooks County officials to provide forensic assistance in the identification and repatriation of these individuals.

RFP began receiving the skeletal remains of undocumented immigrants in March 2013 and began grave exhumations in May with a forensic team from the University of Indianapolis. Field estimation indicated there were 63 individuals exhumed; however, that number has increased during laboratory analysis due to the discovery of body bags containing multiple individuals and some comingling. In addition, personnel from Baylor University make frequent trips to Brooks County to take possession of unburied unidentified remains. The goals of this effort are to provide anthropological analysis, DNA sampling, and database entry for the purpose of identification. As of July 2013, 98 cases have been recovered and are in various stages of analyses. Forty-three cases were transported to Texas State University where they are processed, anthropologically analyzed, DNA sampled, and material goods photographed. In addition, the University of

Indianapolis (n=12) and Joint POW/MIA Accounting Command Central Identification Laboratory (JPAC-CIL) (n=23) have taken cases for anthropological analyses and DNA sampling. The remaining cases are being analyzed at Baylor. All case information, forensic analyses and photographs are submitted to National Missing and Unidentified Persons System (NamUs) and the DNA results entered into Combined DNA Index System (CODIS) by the University of North Texas. Any unidentified remains will be housed at Baylor University and the cases will remain open and active utilizing any new technological breakthroughs that may become available. The exhumation efforts will continue until all unidentified remains are analyzed and fatalities cease. Unfortunately, the death toll for 2013 is similar to that of 2012.

Undocumented Immigrants, Border Deaths, Exhumations

H122 An Integrated Approach to the Identification of Human Remains: The Biocultural Profile of Undocumented Migrants

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After attending this presentation, attendees will learn about the integration of sociocultural and biological factors used to establish the biocultural profile of undocumented migrants at the Pima County Office of the Medical Examiner (PCOME). This presentation will expand upon the Undocumented Border Crosser (UBC) profile and advocate for a biocultural approach to forensic anthropological casework.

This presentation will impact the forensic science community by introducing the biocultural profile of undocumented migrants, which will assist medical examiners' and coroners' offices in the identification of probable Mexican and Central American migrants dying within United States jurisdictions.

Over the past decade, the PCOME has examined the remains of over 2,000 suspected UBC's from various regions of Latin America. Approximately 1,300 of these individuals have been identified as foreign nationals, while the remaining 700 are likely migrants as well. The large caseload and unique circumstance of people dying while crossing the border compelled examiners at the PCOME to create a systematic way of distinguishing between American and foreign-born Hispanics, and lessons learned from the examination of 1,300 identified migrants has led the PCOME to take these criteria and apply them to suspected migrants. The resulting UBC profile distinguishes Hispanic foreign nationals through locational context, personal effects, and an anthropological determination of ancestry consistent with individuals from Latin American countries.¹ The "cultural profile" was designated to describe the non-biological aspect of the UBC profile, including culturally specific personal effects like religious icons, and foreign identification cards, and currency, body modifications such as cosmetic dental ornamentation, and short stature and poor oral health.² Reineke and Anderson argue that "since migrants are defined not by biology or culture but by the fact of marginality—people clandestinely entering the country for economic reasons—each marker in the suite of characteristics that make up the UBC profile should indicate marginality in this border context."³ Therefore, many features of the cultural profile are social or economic in

nature and should be labeled the “sociocultural profile,” since both socioeconomic and cultural factors are assessed.

The first goal of this presentation is to expand upon the sociocultural profile and introduce the “biocultural profile,” a component of analysis that can narrow the range of possible sub-groups in unidentified persons cases. In this context, the biocultural profile is defined as the manifestation of cultural and socioeconomic factors on the physical body, whether integrated into one’s biology (e.g., skeletal indicators of health, oral pathologies) or applied as semi-permanent modifications observable on one’s remains (e.g., tattoos, dental ornamentation, dental restorations). As demonstrated at the PCOME, these features, analyzed in conjunction with the biological assessments of age, sex, ancestry, stature, and increasingly DNA profiles, can further differentiate individuals as belonging to more specific economic, cultural, social, or ethnic groups.⁴

The final goal of this presentation is to present a biocultural profile specific to undocumented migrants, which can be useful in differentiating the remains of undocumented Hispanic migrants from those of American-born Hispanics. Many characteristics of the UBC profile, such as locational context or personal items carried while crossing the border (food/water, foreign identification media, and papers with phone numbers), are not appropriate to migrants living within the United States; however, features of the sociocultural and biocultural profiles (tattoos, cosmetic dental work, and skeletal indicators of stress) are applicable everywhere. Within the undocumented migrant community, a significant component of the biocultural profile includes biological indicators of marginality such as poor oral health, poor dental work, short stature, poorly-set healed fractures, and skeletal indicators of stress, all of which may remain visible on the body long after an individual has migrated to the United States.⁵

With anticipated immigration reform and an ever-expanding Hispanic migrant populace living within the United States, it is increasingly necessary for medical examiners’ offices throughout the country to become familiar with the biological and sociocultural characteristics of undocumented migrant remains. The biocultural profile of undocumented migrants is broadly applicable and can apply to individuals who have been living within the United States as an undocumented and marginalized community.

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Forensic Anthropology, Biocultural Profile, Undocumented Migrants

H123 Initiating the Mexican Water Isoscape: Using Water Isotopes for Region of Origin Identification

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After attending this presentation, attendees will better understand issues surrounding the use of water isotopes for region origin identification in Mexico.

This presentation will impact the forensic science community by presenting data on the relationship between bottled and tap waters and the utility of these water types as predictors of region of origin and/or migration.

Despite a drop in apprehensions since 2006 and the Pew Hispanic Center’s report of zero net migration from Mexico for 2012, the yearly death toll for undocumented migrants along the border region was 477 individuals for 2012.¹ Identification of undocumented populations that reach the U.S is complicated by many factors including the multiplicity of sending areas and the lack of traditional forms of identification or records.

The use of oxygen and hydrogen isotopes in human drinking water, hair, bone, fingernails and teeth has demonstrated the ability to track movement and identify the region of origin for modern populations in the United States, Europe, and Asia.^{2,3} Mexicans have the highest bottle water consumption in the world, drinking four times more bottle water than United States residents.⁴ Bottle water consumption from non-local sources weakens the ability of oxygen and hydrogen isotopes to generate predictive trends identifying region of origin from human tissues.

The purpose of this study is to: (1) explore the relationship between oxygen and hydrogen isotopes in tap water and the various bottled water sources available to Mexican populations; and, (2) Establish the precision with which this relationship can identify region of origin and/or migration within Mexico. The samples consist of 43 samples of bottled water collected from seven states in Central and Southern Mexico and the Federal District, 49 samples of tap water collected from nine states in Central and Southern Mexico and the Federal District, and hair samples collected from 13 separate locations spanning four states. GPS information was recorded for all tap water and hair samples. Purchase location was noted for all bottled water samples and birthplace information was recorded for all tooth samples. Bottled waters came from 15 different companies selling bottle water in Mexico. Hydrogen and oxygen water isotopes were measured for tap water samples and bottled waters samples for all locations using laser absorption spectroscopy at the University of Utah Stable Isotope Ratio Facility for Environmental Research (SIRFER) laboratory. All statistical analysis was completed using SPSS version 20 for Mac®.

All reported data has been calibrated to the Vienna Standard Mean Ocean Water-Standard Light Antarctic Precipitation (VSMOW-SLAP) scale. Bottle water values spanned a range from -11.45‰ to +4.36‰ and -79.79‰ to +26.34 for $\delta^{18}\text{O}$ and $\delta^2\text{H}$, respectively. Tap water values spanned a range from -11.86‰ to -3.90‰ and -83.3‰ to -20.14‰ for $\delta^{18}\text{O}$ and $\delta^2\text{H}$, respectively. The most ^2H and ^{18}O depleted tap water samples were distributed over the inland regions of Mexico City, Puebla, Hidalgo, and Morelos. The $\delta^{18}\text{O}$ and $\delta^2\text{H}$ data were strongly correlated for both tap $r = .987$ and bottled water samples $r = .990$. The local meteoric water line for tap waters was $\delta^2\text{H} = 8.688X \delta^{18}\text{O} + 14.945$ $r^2 = 0.975$. The local meteoric water line for bottle waters was $\delta^2\text{H} = 6.797X \delta^{18}\text{O} - 2.856$ $r^2 = 0.97$. The slope of the LMWL for tap waters is similar to that of the Global Meteoric Water Line (GMWL) which has a slope of 8; however, the LMWL for bottled waters has a slope of ~6. This difference in slope may indicate that the waters included in the bottled water samples originated in regions that were: (1) different or more restricted than the tap water samples; and, (2) were more arid. Independent *t*-test demonstrated a statistically significant difference between mean $\delta^{18}\text{O}$ values between tap and bottled waters $t(61) = -3.007$, $p = 0.004$. The $\delta^{18}\text{O}$ mean value for bottle waters was -9.5‰ , which was on average $1.93\text{‰} \pm .65\text{‰}$

more depleted in ^{18}O than tap water values, supporting the previous assertions. Region of origin for hair data were analyzed using the predictive model developed by Ehleringer *et al.* for United States populations. An independent *t*-test confirmed that the generated values for source tap waters were not significantly different from the mean values for source location $p=0.001$. Due to the small sample size of this preliminary work caution should be taken in the interpretation; however, results suggest that Mexican tap water can be used to predict Mexican region origin based on hair samples.

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Water Isotopes, Region of Origin, Mexico

H124 A Structured Approach to Assessing Morphogenetic Variation in Mexico: Tests of Method Informedness for Improved Skeletal Identifications

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After attending this presentation, attendees will have a clearer understanding of population structure at a smaller geographic scale within Mexico. Additionally, attendees will learn how various statistical approaches applied to estimate population structure influence the conclusions drawn about biological patterning.

This presentation will impact the forensic science community by ameliorating the immediate need to improve skeletal identification methods for U.S.-Mexico border-crossing fatalities through a fine-scale approach to inferring skeletal and DNA variation across Mexico. By determining how heterogeneity within Mexico is expressed through differences in skeletal morphology and genetic profiles, forensic anthropologists are able to make informed decisions about method development and best practices for human identification contexts.

Population structure within Latin America has been well documented in previous studies. A clearer understanding of structure at a smaller geographic scale is a critical next step. Revealing latent structure that exists within morphogenetic data, and establishing its correspondence to geography, can improve forensic anthropological methods for estimating the biological profile. By demonstrating that Hispanic populations are biologically heterogeneous and determining how this variation is expressed through differences in skeletal morphology and genetic profiles, informed decisions can be made about method development in forensic anthropology and best practices for human identification contexts. The immediate need to improve skeletal identification methods for U.S.-Mexico border fatalities demands that such a

fine-scale approach is applied to morphogenetic variation within Mexico.

Preliminary studies of both genetic and craniometric data demonstrate that population structure is detectable within Mexico and conforms to known geographic boundaries of population settlements and gene flow.^{1,2} Building on these recent observations, it is argued that patterning within Mexico itself may confound estimations of the biological profile; thus, further investigation into the effects of geography on morphogenetic variation can improve forensic methods by ensuring appropriate targeting and application. To this end, the present study examines how using various statistical approaches to estimate population structure influences the conclusions drawn about biological patterning. This study tests multiple strategies for inferring structure for genetic Short Tandem Repeat (STR) and cranial Incongruence Length Difference (ILD) data, each strategy with differing levels of *a priori* model informedness, using cases from the Pima County Office of the Medical Examiner. Approximately one-third of the 300+ sampled cases were identified individuals from Mexico. Using their state of origin, the study tested how different statistical approaches convey structure within Mexico and evaluated how well these inferred groupings correspond to geography.

Three tiers of model informedness were tested: low- (LLI); mid- (MLI); and full- (FLI) level informedness. LLI models were assayed using unsupervised model-based clustering, implemented with MCLUST and Structure 2.3.1 software, for ILD and STR datasets, respectively. Components and model parameterization were determined for the LLI analyses in the absence of any known identifiers. For each case, STR or ILD data was input, while region of origin was excluded; the biological data alone drove the clustering. For the MLI-level analyses, structure was assessed for ILD and STR data using k-means clustering. While region of origin data was excluded, the cluster-seed approach was designed to maximize the biological data variation among clusters. Finally, the FLI tests included a mixture model-based discriminant function analysis for ILD data, and a Structure 2.3.1 analysis, following the recommendations of Hubisz *et al.*, for STR data.^{3,4} For both FLI models, region of origin data was combined with the biological data for cluster partitioning, thus making these models fully "informed."

The resulting clustering for the LLI, MLI, and FLI approaches revealed a clear trend: increased informedness yields increased correspondence with geography and between cluster memberships from ILD and STR data. ILL models showed no significant indications of population structure for either ILD or STR data (when $K \geq 2$). In contrast, both MLI and FLI approaches yielded geographically structured clusters, with the FLI outperforming the MLI. The results of this study demonstrate that sub-population structure can be detected using ILD and STR data from Mexico and that the level of method informedness greatly affects its ability to convey this structure. In the case of Mexico, where variation is regional and differences, while present, are nuanced, it is critical researchers consider adequate samples and select the most appropriately informed models.

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Mexico, Population Structure, Regional Variation

H125 The Postmortem Interval: A Retrospective Study in Desert Open-Air Environments

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After attending this presentation, attendees will better understand the escalation of migrant deaths along the U.S.-Mexico border and the need to re-examine decomposition rates for human remains recovered from open-air desert environments. This study builds upon previous research by Galloway *et al.* (1989) and contributes an analysis of the postmortem interval for outdoor deaths in the American Southwest that accounts for seasonality, altitude, and precipitation.¹

This presentation will impact the forensic science community by providing a predictive value by which to estimate Postmortem Interval (PMI) in cases of death within an open-air desert environment in seasons of varying temperature and rainfall.

Forensic anthropologists at the Pima County Office of the Medical Examiner (PCOME) in Tucson, Arizona, are tasked with approximately 150 cases per year. Migrants dying in the desert are a prominent component of this caseload and current trends indicate this is increasing border-wide. Many of these remains are not readily identifiable due to quick decomposition in the desert, often further complicated by migrants' efforts to obscure their identities. In these cases, estimating PMI is an important albeit challenging endeavor, as most studies are problematic due to regional differences in climate, as well as complexities in research design such as small sample sizes or the use of pigs as human proxies. Currently, the only relevant PMI estimation method for use within the southwest region is a study by Galloway *et al.*, which uses a retrospective analysis of cases from the PCOME to estimate decay rates of human remains in southern Arizona. While the study addresses a variety of circumstances, only 51% of the cases were found in open-air environments and the sample of individuals within the upper ranges of decay was quite small, due to difficulties in identifying highly degraded remains in the 1980s.

This presentation intends to address these issues by conducting another retrospective study using positively identified individuals who died within an open-air desert setting and had last known alive and recovery dates. In 2011, a decomposition scoring system was devised to classify remains at the PCOME into several categories: (1) fresh; (2) decomposed; (3) decomposed with focal skeletonization; (4) skeletonization with soft tissue mummification; (5) skeletonization with articulation/ligamentous attachment; (6) complete skeletonization with disarticulation; and, (7) skeletonization with bony degradation. These decomposition categories were ascribed to scene and autopsy photographs of 156 identified decedents at the PCOME. For each case, the time-since-death was calculated by the total number of days between last-known-alive and recovery dates. Conditions such as shade, direct sunlight, location within a wash, precipitation, ambient temperature, and season of recovery were noted. Recovery

locations were mapped with the Arizona OpenGIS Initiative for Deceased Migrants to record altitude and information about the surrounding topographic environment.² Climatological data was retrieved from National Oceanic and Atmospheric Administrations weather stations.

The results of this study confirm Galloway and colleagues' findings for early decomposition categories, but demonstrate a somewhat accelerated initial skeletonization process. Fresh remains are typically recovered within one week of death and decomposition occurs within a month, with temperature and seasonality playing a large role in the speed of decay. Mummification ranges from 6 to 145 days after death, tends to delay the usual decomposition process and, as Galloway *et al.* point out, is linked to drier months with little precipitation. Focal skeletonization can occur in just a week or two, especially during the rainy monsoon season. In contrast to Galloway *et al.*'s results, this study indicates that skeletonization with articulation/ligamentous attachment can occur as early as 17 days, and skeletonization with disarticulation is seen as early as 39 days in the hot summer monsoon months. For individuals exposed to sunlight, bleaching can occur in just a few months; however, bony degradation to include longitudinal cracking and cortical exfoliation does not typically occur until after 1.5 years. The skeletonization process is quite variable and is likely tied to multiple factors including precipitation, exposure to direct sunlight, and animal scavenging. This research supplements Galloway *et al.*'s study by expanding the sample size of individuals found in open-air settings and provides a standardized means to estimate postmortem interval in the southwestern desert and similar arid environments.

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Postmortem Interval, Open-Air Desert, Decomposition

H126 Life After Human Death: The Thanatobiome

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After attending this presentation, attendees will understand the meaning of the word "thanatobiome" and the new field of study of which little is known.

This presentation will impact the forensic science community by increasing understanding of the microbiology of decaying human bodies and provide additional information to those who are interested in a new possible way to determine Postmortem Interval (PMI) based on the human microbiome.

A paradigm shift in our understanding of cells in the human body has occurred as a result of human microbiome studies. Consider this: before birth, all cells in a baby's body are human. This is the only stage of development that humans are essentially "microbial-cell free." When a baby passes through the uterus, consumes food, and interacts with other humans and the environment, it becomes inoculated with microbes that live and thrive in its body. One of the truly amazing findings of the human microbiome studies is that, in adulthood, most cells in a human body are not human at all—rather, 90% are microbes. This means that microbes are very much a part of "us" and implies that they

play important roles in both our health and disease. Given these facts, the study postulated the role of the microbiome after a human dies. To date, all human microbiome studies have focused on the microbes of living beings. Amazingly, almost nothing is known about the composition and abundance of the human microbiome after death. Here are the sparse details we do know: (1) microbial cells proliferate in the body after death because the immune system ceases to function and there is a massive release of nutrients from human cells that lose membrane integrity; (2) the microbial proliferation is time/temperature-dependent, and begins at the ileocecal area, spreads to the spleen and liver, and eventually the heart and brain; and, (3) microbes colonize the ileocecal area by invading capillaries of the lymphatic and vascular systems and, in the case of the respiratory system, by invading the mucus membranes.

Based on this information, the following working hypothesis was formulated: the composition and abundance of microbes in the human thanatomicrobiome (i.e., *death*-microbiome) varies by organ and changes as a function of time and temperature. This hypothesis is relevant to the determination of PMI because the abundance of certain microbes in different organs could be indicative of the elapse time since death. This hypothesis is also relevant to human evolution studies because, just as microbes have evolved in human development, it is likely that they have evolved with us in human death. To test the hypothesis, the thanatomicrobiome in blood, liver, spleen, heart and brain samples of human cadavers having a range of known PMIs was surveyed. Surveying the thanatomicrobiome was accomplished by extracting DNA from the organs, amplifying the 16S ribosomal RNA genes, and sequencing the genes on a 454 pyrosequencer. The obtained sequences were uploaded to MG-RAST for annotation and determination of the microbial abundances at different levels of taxonomic resolution. Because the PMI and temperature conditions for each cadaver are known, the thanatomicrobiome in different organs will be determined and related to the time of death. It is thought that this is the first study of the human thanatomicrobiome to use DNA sequencing technology.

Thantomicrobiome, Postmortem Interval, DNA Sequencing

H127 Refining Postmortem Interval Estimates in the Northeast

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After attending this presentation, attendees will be familiar with new outdoor scene data-collection protocols used to improve postmortem interval estimates for human remains found in the woods of Pennsylvania and the Northeast.

This presentation will impact the forensic science community by standardizing data collection protocols at outdoor body recovery scenes that will result in significant improvements in the estimation of postmortem intervals, critical to forensic investigations of unidentified remains.

When human remains are found at outdoor forensic scenes, reliable assessments of Postmortem Interval (PMI) become a critical component of forensic investigations. During the first hours or days after the death, forensic pathological indicators of decomposition provide the best estimates. However, after that initial time frame, the forensic anthropologist is expected to provide reliable estimates based on taphonomic analysis. During the past few decades, experimental studies have served to improve

our knowledge on the subject, although serious limitations to the sample sizes and environmental regimes at which comprehensive studies can be carried out still remain.

Case studies, therefore, become especially relevant to guide and complement experimental studies. Most PMI estimates rely largely on the forensic anthropologist's personal case experiences in very specific geographical areas. A major problem is that case studies have rarely been compared and systematized. This is partly explained by the fact that in order to take into account the multiple taphonomic factors affecting the remains, scientific estimates of PMI must include detailed consideration of the context in which the remains were found. Collecting the necessary contextual data is only possible when the scene is processed through forensic archaeological techniques, still a rather uncommon occurrence.

This study proposes that these problems are similar to those faced by clinical trials for diseases with low incident rates, in which no single hospital receives enough patients to provide an appropriate sample size and, consequently, it is necessary to share and combine data from different healthcare facilities. The key in these situations is the standardization of treatment, research, data collection, and coding protocols employed by multiple institutions.

In this study, a series of case studies compiled during the past two decades are presented, paying particular attention to the types of contextual data collected and the techniques used to collect them. Discussion includes their potential utility for obtaining reliable PMI estimates in Pennsylvania and surrounding environs.

Mercyhurst University forensic archaeological teams have been documenting outdoor crime scenes from a forensic taphonomic perspective for nearly 20 years. A previous presentation in 1995 (Dirkmaat and Sienicki 1995) provided a basic outline for determining PMI in Pennsylvania based on the analysis of 15 outdoor forensic scenes. It was determined from this study that even after a full summer outside, some soft tissue remains associated with the bones. At least two summers on the surface were required to remove all soft tissue.

The current study expands this analysis to include nearly 40 additional outdoor forensic cases of known PMI from three states (Pennsylvania, New York, and Ohio) since 1995. Discussed are not only the cases themselves, but the different layers of information added to documentation protocols, especially the recordation and measurement of numerous taphonomic factors such as burial vs. surface scatter, the role of scavengers on the rate of decomposition, the amount of shade, site slope, and others. Detailed hand-drawn plan view and profile maps of these scenes allow for the analysis of scattering patterns and other spatial distribution of evidence factors. Geographic Information Systems (GIS) software was used to collect data at larger scales in order to include such factors as local topography, site altitude, soils, tree cover, and others.

These field data were complemented with laboratory documentation and photographs of the condition of the soft tissue by body region. Factors that play a part in decomposition regimes include the amount and location of clothing and personal effects. This data collection strategy permitted the fine-tuning of PMI estimates to include the role of season or month of initial deposition of the body, among other factors. It is proposed that the exchange and discussion of similar protocols and information among forensic archaeology teams can play a critical role in advancing our understanding of decomposition rates and ultimately improve PMI estimates in a variety of different regions and under different conditions.

Postmortem Interval Estimates, Forensic Taphonomy, Forensic Archaeology

H128 The Stone Cold Truth: Preliminary Results of the Effect of Concrete Encasement on the Rate and Pattern of Soft Tissue Decomposition

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After attending this presentation, attendees will gain knowledge of the rate and pattern of decomposition of remains interred in concrete over one year in southern Illinois. Furthermore, attendees will learn the best practice for removing remains from concrete while maintaining the highest level of evidence preservation.

This presentation will impact the forensic science community by reporting a successful method for removal of remains encased in concrete and highlighting differences in the rate and pattern of individuals encased in concrete when compared to individuals decomposing in other taphonomic circumstances. The results of this study will show that estimates of Postmortem Interval (PMI) using traditional methods differ dramatically when used on "natural" and concrete interment victims, possibly hindering several aspects of criminal investigation.

Eight pigs (*Sus scrofa*) (27-36kg) were assessed to investigate differences in decomposition between individuals buried in concrete versus surface placement. All research in this study took place at the Complex for Forensic Anthropology Research (CFAR). Subjects were attained from the Southern Illinois University (SIU) Swine Research Center and were euthanized for other reasons of individual health. Each subject was placed on a 15cm bed of Quikrete Concrete® in a one-meter-square hole dug 30cm deeper than individual laying height. Enough concrete was then poured over the subject to cover it with an even depth of 15cm of concrete. Depending on the size of the pig, this required 437-582kg of concrete (11-15 80lb. bags). A control subject was placed on the surface at CFAR at the same time as initial encasement.

Subjects were removed from their encasements at monthly intervals for five months and bimonthly intervals from the next six to twelve months. Methods of removal ranged from using manual concrete chisels and hammers of various weights, to air, and electric, powered chisels. The best method identified in this study involved digging ~0.5m-wide trenches around the interment for ease of access to the sides of the concrete block and then removing 2.5cm strips of concrete with pointed concrete hand chisels. Care must be taken to ensure soft tissue and material evidence attached to the concrete is retained.

Results demonstrate encasement in concrete severely slows decomposition, with subjects excavated after one year being in early stages of decomposition. The Total Body Score (TBS) method was utilized at the time of excavation for each subject to assess decomposition stage.¹ The concrete subjects yielded substantially lower TBS. By three months (25,949.45 Kelvin Accumulated Degree Days (KADD)), the control subject was nearly completely skeletonized yielding a TBS of 30, while the concrete subject yielded a score of 7, barely entering the "early" decomposition stage. Skin slippage of concrete subjects was first witnessed at four months. At one year (105,339.16 KADD) purging of fluids and intact organs were present yielding a TBS of 12 for the final subject excavated. These results are contrary to recent findings of Gibelli and colleagues, who report mummification,

significant adipocere formation, and partial skeletonization of subjects after six months in concrete.²

Additionally, decomposition pattern differed greatly between control and research subjects. Individuals encased in concrete may be more difficult to locate in clandestine disposal situations, as no odor permeated the concrete until it was breached, insect activity was not noticeably higher in the local area, and motion-activated cameras revealed no scavenger interest in these subjects. The outer layer of skin was also chemically burned and adhered to the concrete. This was most likely due to both the exothermic reaction of the concrete curing process (reaching temperatures over 60°C) and exposure to harmful chemicals in the concrete such as lime and calcium sulfate. Any superficial markings or staining on the skin of the victim may be compromised due to these processes, so examination of these concrete pieces should be performed to minimize evidence loss.

The recorded data show differences in both the rate and pattern of decomposition when subjects interred in concrete are compared to controls placed on the ground surface. Concrete encasement protects subjects from scavengers and insects alike, severely hindering decomposition. The pattern is also unique, as the outer layer of skin burns and adheres to the concrete. One should expect traditional methods of estimating PMI to yield skewed results when dealing with concrete interments. Continued research is necessary and ongoing.

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Concrete Burial, Taphonomy, Forensic Anthropology

H129 Raccoon Scavenging at the Anthropological Research Facility at the University of Tennessee, Knoxville

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After attending this presentation, attendees will appreciate raccoon scavenging as documented by occurrences within the Anthropological Research Facility (ARF) at the University of Tennessee, Knoxville (UTK) and, therefore, better understand their impact on the decomposition of human remains.

This presentation will impact the forensic science community by demonstrating seasonal and annual effects of raccoon scavenging on human cadavers. In addition, this research demonstrates that scavenging by raccoons is more likely to cause mummification in human remains, thus leading to increased confusion in some forensic anthropological issues such as estimation of a postmortem interval.

Although raccoons (*Procyon lotor*) are one of the most common scavengers found across the United States, studies regarding their scavenging patterns on human remains have been limited. Moreover, their forensic anthropological significance has not been well appreciated. The goal of this research is to explore temporal patterns of raccoon scavenging on human cadavers, while emphasizing its implications in forensic investigations.

Daily taphonomic changes of donated human cadavers at the ARF were observed through the daily photo database of the Forensic Anthropology Center at UTK. A total of 118 cadavers, which were placed at the ARF between February 2011 and December 2012, were examined to see if raccoon scavenging occurred during decomposition. Damage due to raccoon scavenging was determined using demonstrations by Synstelien as well as empirical knowledge of raccoon scavenging patterns the presenter (Jeong) achieved while working at the ARF for years.¹

Of the 118 cadavers, 57 (48.3%) were scavenged. Although the annual difference in scavenging frequency between 2011 and 2012 was marginal (49% and 47.8%, respectively), seasonal differences were obvious. Overall scavenging frequency during "hot" months (i.e., May to September), when monthly temperatures were above 20°C, was much higher (62.5%) than that of cooler months (35.5%). For cadavers that were placed during the hottest month of July, scavenging frequency was at its highest (83.8%).

Initially, raccoon scavenging began while cadavers still retained muscle tissue in their limbs. More than half (63.2%) were scavenged when they were in the fresh or skin slippage stage, while only 3.5% were scavenged in the advanced decay stage. Calculation of the days between the cadavers' placement at the ARF and the time of initial scavenging showed an obvious seasonal pattern. After placement during the "hot" months, raccoons began to scavenge an average of 5.9 days later whereas during the cooler months, initial scavenging averaged 16.3 days after placement.

A similar seasonal pattern was observed with regard to the duration of scavenging. The length of time raccoons continued to scavenge was significantly shorter during "hot" months, averaging 7.6 days yet continued in cooler months on average for 28 days. In terms of annual differences, scavenging on cadavers occurred longer in 2012 than in 2011.

It was also observed that scavenged cadavers resulted in mummification, while unscavenged cadavers tended to be skeletonized. This demonstrates that a significant difference in the rate of mummification exists between scavenged (82.5%) and unscavenged (34.4%) remains ($\chi^2=28.40, p<0.001$).

This research describes and quantifies scavenging patterns of raccoons. Although some issues such as the cause of annual differences in initial scavenging, scavenging duration, and the mechanism for mummification by scavenging, could not be clearly demonstrated, raccoons can be said to be associated to and impact the final stage of decomposition (i.e., mummification or skeletonization). The significance of raccoon activity in terms of forensic investigation should be considered with more research in the field of forensic anthropology.

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Raccoon, Scavenging, Human Cadavers

H130 Methods for Recognizing, Collecting, and Analyzing Vulture Evidence in Forensic Contexts Obtained Through GPS Tracking and Long-Term Scavenging Research

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After attending this presentation, attendees will be aware of the 14 skeletal and contextual traits indicative of prior vulture scavenging and how such data can be used to more accurately

estimate the Postmortem Interval (PMI). Additional data will be provided regarding methods for determining vulture scavenging from other sources of skeletal trauma during laboratory analysis of the remains.

This presentation will impact the forensic science community by providing new criteria for recognizing vulture-scavenged remains and for differentiating the skeletal marks caused by vultures from those left by other animal scavengers as well as the peri-mortem trauma inflicted by humans. Furthermore, this is the first forensic study to use satellite telemetry, or animal Global Positioning System (GPS) tracking, to help explain the taphonomic processes of vulture scavenging.

Vultures are the world's only obligate scavengers and are known to scavenge humans and accelerate decomposition, yet methods for recognizing, collecting, and analyzing vulture evidence from forensic contexts do not exist.¹⁻³ To address this problem, vulture taphonomic data were collected from controlled decomposition studies as well as from 10 months of trapping and GPS tagging of vultures at the Texas State Forensic Anthropology Research Facility (FARF) and surrounding areas.

Over a two-year period, 43 uncaged juvenile pigs were placed across 6,000 km², dozens of animals were used to bait a vulture trap, and 52 vultures were housed and monitored for 10 days. Results from these studies, combined with the ongoing hourly monitoring of five vultures fitted with 70-g solar Argos/GPS PTT-100 tracking devices, provide comprehensive insight into how vultures scavenge and reveal that vulture scavenging is influenced largely by the birds' physiology and their need to liftoff and soar post-scavenging.

These physiological limits of vultures appear taphonomically. Vultures often began scavenging by pulling the internal organs and soft tissues out and away from the ribs and spine, leaving the heavy and less nutritious vertebral column behind. A total of 14 taphonomic characteristics were observed in relation to vulture scavenging, and each is related to the biology and soaring behavior of the bird. The key traits for recognizing vulture scavenging are an intact vertebral column and down (plumulaceous) feathers left at the site. The down feathers can be used to both identify the species of bird and more accurately estimate the PMI.⁴

Although variation does exist in vulture scavenging behavior both over time and between different-aged vultures, certain traits indicative of vulture scavenging were present regardless of the type of bait being scavenged. Reoccurring similarities were observed in the vulture scavenged juvenile pigs, the animals used to bait the vulture trap, and the human cadaver scavenged by vultures and depicted in the 2012 presentation by Mavroudas and colleagues.⁵

The results from this study will allow investigators to make more informed decisions about when to include vulture scavenging into assessments of the crime scene, PMI estimations, and trauma analysis associated with unidentified human remains. Vultures accelerate decomposition, so if their scavenging is unaccounted for, then PMI estimates may suggest an overestimate in the amount of time between death and discovery, and the marks left by vultures may be misinterpreted as peri-mortem trauma inflicted by a human.

Support for this research was provided by the National Science Foundation (NSF) Doctoral Dissertation Research Improvement Grant (PD 98-1352) and the Louisiana State University (LSU) West-Russell Award. The findings and opinions to be presented are those of the author and not necessarily those of either the NSF or LSU.

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Vulture Scavenging, Taphonomy, GPS Tracking

H131 Differences in Soft Tissue Decomposition Rate and Manner Between Previously Frozen and Never Frozen Domestic Pigs (*Sus Scrofa*)

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After attending this presentation, attendees will understand the effects of previous freezing on the progression of both internal and external soft tissue decomposition.

This presentation will impact the forensic science community by contributing to a greater understanding of the variables affecting decomposition as well as providing a suite of characteristics that warrant consideration of exposure to freezing in forensic cases.

This research examined differences in decomposition rate and manner of domestic pig subjects (*Sus scrofa*) in never-frozen (control) and previously frozen (experimental) research conditions. Due to the detrimental effects of freezing on bacteria responsible for putrefaction and previous taphonomic studies concerning the effects of freezing on rat models and pig skeletal muscle, it was hypothesized that experimental subjects would undergo less putrefaction and decompose at a slower rate than control subjects.¹⁻³

Eight control subjects and eight experimental subjects were weight-matched and placed in an outdoor research environment within an hour of each other on the same day during two seasons (fall and winter) at the Complex for Forensic Anthropology Research (CFAR) at Southern Illinois University (SIU). All subjects were obtained from the SIU Swine Research Center and were euthanized due to health issues. No subject was euthanized for the purpose of this research. Subject pairs were placed 1-2m apart in identical environmental circumstances and protected from scavengers under a wire mesh cage. Subjects selected for the experimental condition were frozen at -18°C for a minimum of seven days. Subjects weighed at least 23kg, and weight difference within pairs was minimized (<15%) to ensure observed differences in decomposition were due to the research condition being investigated. Extensive quantitative observations were collected daily: abdominal circumference; Total Body Score (TBS); and temperature.⁴ Qualitative observations were also recorded daily: photographs of subjects; descriptive decomposition stages; and general visual observations concerning insect activity and subject appearance.⁵ Field necropsies, consisting of qualitative macroscopic observations of the abdominal and thoracic organs, were performed at varying Accumulated Degree Days (ADD) ranging between 50 and 300 (Celsius). Comparison (paired samples *t*-tests) of ADD to three TBS (early decomposition TBS,

9.5 TBS, and advanced decomposition TBS) indicated the rate of decomposition of experimental subjects was significantly slower than control subjects at both early decomposition TBS and 9.5 TBS ($p=0.003$ & $p=0.002$, respectively). In experimental subjects at these two stages, thermal energy was directed toward thaw instead of to decomposition. This, in concert with decreased viable putrefactive bacteria, resulted in a slowed rate of decomposition. However, no difference in ADD required to reach advanced decomposition was observed between the two conditions ($p=0.112$). Only eight subjects reached the advanced decomposition stage; thus, further research is necessary and ongoing for subjects at later stages of decomposition.

Differences in qualitative observations between experimental and control subjects were also noteworthy. Internally, experimental subjects displayed less internal putrefaction, limited internal color change, minimal organ distention, and dehydration of the heart. In contrast, control subjects exhibited increased putrefaction, more internal color change, substantial organ distention, and pleural bubbling around the lungs. Externally, experimental subjects displayed overall gray discoloration, increased desiccation, more oviposition by insects, and nearly non-existent bloat. Externally, control subjects exhibited normal color change during decomposition, with purple and green discoloration being much more common in these subjects. Bloat was extensive in control subjects. The mean of the percent difference in abdominal circumference between experimental and control subjects was significant (control mean = 1.07, experimental mean = 1.00, paired samples *t*-test, $p<0.001$). Due to these differences in rate and manner between experimental and control subjects, this study suggests previously frozen subjects should not be used in taphonomic research, as results may not accurately reflect the "normal" taphonomic condition. In cases of forensic significance, the possibility of freezing should be investigated if the victim displays external gray discoloration, substantial external desiccation, decreased internal putrefaction, minimal organ distention, lack of bloat, and a dehydrated-appearing heart.

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Freezing, Forensic Taphonomy, Decomposition

H132 Scavenging of Human Remains Within a Human Decomposition Research Facility in Western North Carolina

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After attending this presentation, attendees will have a better understanding of scavenging activity and the impact of these scavengers on human decomposition within an outdoor human decomposition research facility located in rural western North Carolina. The goal of this research is to identify mammalian species that may influence the decomposition process. The influence these species may have on one another will be discussed.

This presentation will impact the forensic science community by examining the role that avian and scavenging mammalian species play on the decomposition process and will broaden the scope of existing knowledge of scavenging of human remains within a secured human decomposition facility.

Our understanding of the decomposition process is limited by the small number and scattered locations of human decomposition research facilities in the United States. Reporting the results of small-scale studies such as this one is an important step in understanding broader patterns of decomposition phenomena as well as regional differences. Such studies are also important in generating additional hypotheses to be tested. This research focuses on the role that certain species of scavengers play in the decomposition process within a controlled environment. These data are of importance to pathologists and physical anthropologists when tasked with estimation of postmortem interval and interpretation of scattered remains.

Over a period of seventeen months (June 2011-October 2012), six sets of human remains that had been placed on the ground surface within an outdoor human decomposition research facility in the mountains of western North Carolina were observed for decompositional changes. Motion-activated game cameras were utilized to track scavenger activity, and field visits were conducted to track changes in the remains.

This research resulted in the identification of "repetitive" and "sporadic" scavengers of human remains within the confines of the outdoor human decomposition facility in western North Carolina. When compared to previous scavenging studies completed at other human decomposition research facilities, the scavengers identified for this locale were similar. The "repetitive" scavenger was identified via consistent and regular appearances and feedings throughout the observation period. Although the "sporadic" scavengers may have consumed more at individual feedings, their appearances were not as frequent or as consistent as those of the "repetitive" scavengers. The animals identified as "sporadic" scavengers were not the species expected. In particular, the role of avian scavenging was highly diminished in comparison to a previous scavenging study completed within the same location.¹ In addition, it was noted that scavenger interest in the remains varied, the cause of which will need to be explored through future research.

This research identifies several avian and scavenging mammalian scavengers of human soft tissue in the Blue Ridge physiographic zone of western North Carolina. As similar studies are conducted in other regions, our overall knowledge of scavenging of human remains will expand. By becoming more familiar with the results of such studies, professionals may experience a heightened awareness of the presence and activity of scavengers even in a seemingly protected environment.

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Scavenging, Human Remains, Decomposition

H133 Assessment of Hair Lesions Due to Sharp Force Wounds and Entomological Activity: An Experimental Study

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After attending this presentation, attendees will gain knowledge concerning the importance of an accurate analysis of hair in forensic cases to highlight possible signs of lesions which may provide information on the *modus operandi* of a criminal.

This presentation will impact the forensic science community by providing a new perspective concerning the assessment of sharp force lesions on hair and the differential diagnosis with taphonomical artifacts and blunt force trauma.

The analysis of hair from human remains is rarely performed but may provide useful information for the correct evaluation of forensic cases. In detail, hair can be affected by lesions which may be caused by trauma or insects. In addition, finding signs of sharp weapons on hair may provide important information for the reconstruction of the manner of death or may suggest forensic practise. This study stemmed from an actual judicial case where the correct interpretation of hair lesions on locks found on human skeletal remains was crucial for attributing the crime to a specific person who used to cut the hair of his victims (in this case, the defense sustained that the hair lesions present were due to taphonomical or entomological activity).

To study the manner in which hair reacts to blunt, sharp and taphonomical trauma, two different experiments were performed: locks of hair taken from living individuals of different ages underwent lesions by blunt and sharp trauma (both scissors and knife) to verify the morphology of lesions in comparison with those produced by two species of insects (*Tineola Bisselliella* Lepidoptera, Tineidae and *Anthrenus sp.* Coleoptera, Dermestida). Furthermore, dry degraded hair locks from 2- to 6-year-old forensic cases and from ancient 16th-century skeletons were subjected to mechanical trauma in order to verify how degraded hair breaks and whether it can mimic clean cuts. Both locks and single hair, particularly the cut or broken margin, were examined from a macroscopical and microscopical point of view by using stereomicroscopy and scanning electron microscopy.

Results showed the enormous amount of information which can be retrieved from the microscopic analysis of hair: sharp force trauma produces lesions with sharp and regular edges or with broken but “exploded” margins which are still distinguishable from blunt trauma, whereas insects leave single concave lesions. Hair cut with scissors can frequently be distinguished from hair cut with a knife. Dry, brittle hair, on the other hand, breaks with different patterns and is frequently empty inside. This study highlights the enormous amount of information that can be gained from hair found in the context of human remains.

Forensic Anthropology, Hair Analysis, Trauma

H134 The Prodigal Child Returns: The Application of Forensic Taphonomic Analyses to Paleoanthropological Assemblages — Case Examples From the Malapa Hominin Deposit

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After attending this presentation, attendees will obtain knowledge on the application of whole-body forensic taphonomic analyses (focussing on subterranean weathering patterns, insect-bone modification, skeletal disarticulation patterns, and postmortem and post-fossilisation breakage patterns) of two exceptionally well-preserved early hominins. The data presented are used to inform forensic scientists as to the application of analyses of forensically typical, short-duration events into the deep geological past.

This presentation will impact the forensic science community by considering whether or not forensic taphonomy and anthropology can escape from the medicolegal confines of the recent past and provide new perspectives in paleoanthropology and archaeology. On the basis of recent work at Malapa, the answer has to be yes.

The co-option of taphonomy into the forensic sphere has been described by Dirkmaat and colleagues as “the most significant development [to] alter the field of forensic anthropology” in the last 20 years.¹ This paper will argue that forensic taphonomy is now well situated to return the favor by providing a framework with which to investigate the decompositional and formational histories of extremely ancient deposits. This study uses the example of recent analyses applied to a 1.98 million-year-old early hominin assemblage from Malapa, South Africa, the type site of *Australopithecus sediba*.²

Traditional approaches to taphonomic analyses focus on the application of uniformitarian assumptions in order to understand pattern, process, and mechanism in the past. Such studies were often established to answer or address research problems related to early hominid behaviors, such as use of fire or

hunting versus scavenging niches. This early work represents a corpus of literature relating to aspects of necrology, biostratigraphy, burial, and diagenesis of long-dead organisms. In contrast, the epistemological basis of forensic taphonomy is unique in that the field marks a shift in the temporal nature of taphonomic studies, away from longitudinal studies of complex time-averaged assemblages, to shorter postmortem time frames spanning days to years, with the emergence of the individual cadaver as a key unit of analysis.³

In the case of Malapa, two exceptionally well-preserved individuals were recovered from a 1.98 million-year-old karstic deposit in the Cradle of Humanity World Heritage Site, South Africa. MH1 is a juvenile male and MH2 an adult female. The hominins were analyzed initially using classical palaeo-taphonomic approaches, particularly with respect to the interpretation of patterns of peri-mortem breakage and burial rate. However, the later application of an explicit forensic analytical tool kit has seen a rapid re-evaluation of the history of the initial and later postmortem periods in particular. Forensic analyses focussed on macroscopic and microscopic evaluation of breakage patterns, bone surface modifying agents (matrix-based, physical, chemical, insect and invertebrate, mycological, and environmental), the interpretation of decomposition rate and environment based on skeletal part representation and spatial juxtaposition, and pattern of sedimentary loading based on postmortem and post-fossilisation damage in conjunction with fracture-plane analyses of calcified breccia. In particular, analyses of bone surface modification and breakage patterns using explicit biomechanical criteria (which are not conventionally applied in paleontological or archaeological analyses) has resulted in a parsimonious interpretation of the peri-mortem and later depositional history of the two hominin bodies; in particular, the differential pattern of weathering and surface modification between the two individuals suggests that their deposition may have been two separate events, or that micro-environmental conditions existed expressing differing patterns and possible rates of decomposition between the individuals. This study presents recent results, with the emphasis on data derived from macroscopic and microscopic analysis of surface modification and high-resolution spatial analysis of skeletal elements and their sedimentary context from micro-computed tomography and synchrotron imaging. Results indicate: (1) biomechanically-cogent patterns of peri-mortem trauma consistent with low-energy impact; (2) differential patterns of weathering, with MH2 ventrally-focused indicative of final disposition, and MH1 exhibiting a complex pattern of both dorsal and ventral weathering possibly indicative of aerial and sub-aerial processes; (3) widespread invertebrate modification of bone surfaces, including snail and termite damage; (4) contiguous postmortem fractures indicative of sediment-loading consistent with movement of encapsulating matrix; and, (5) spatial context of skeletal elements correlated with high-order rank disarticulation sequence, suggestive of mummification or rapid fixation within the cave system. The analysis of the Malapa assemblage is considered unique, in explicitly applying forensic taphonomic analyses to well-preserved fossilised remains in order to answer what may be considered conventional forensic questions.

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Forensic Taphonomy, Bone Modification, *Australopithecus Sediba*

H135 An Analysis of Process Signatures to Burn Progression of Human Bodies During Commercial Cremation

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After attending this presentation, attendees will have a better understanding of the expected process signatures of burn destruction of bone in a commercial crematory environment.

This presentation will impact the forensic science community by demonstrating that burning is predictable and a predictable progression of thermal destruction to bone is visible and quantifiable.

Distinguishing between thermal damage and non-thermal peri-mortem bone trauma is critical to the forensic analysis of burned human remains. However, standards for such determination developed through the study of actual human remains are limited. Symes *et al.* provides the most detailed analysis of thermal patterns of destruction to the body based on forensic casework.¹ Through this analysis, several process signatures were identified, such as the shielding of soft tissue due to body position, and pugilistic posture of the limbs. Previous bone research frequently relied on pig carcasses (*Sus scrofa*) as proxy cadavers. However, their bones present a distinctly different anatomical structure disallowing a clear comparison to the effects of fire on human anatomy. Performing actualistic studies using human bodies is necessary to fully understand these processes. Past studies into burned human remains have most often focused on one area of the body such as the cranial bones.² Few studies have been conducted using fully articulated bodies. Although Bohnert *et al.* (2008) studied cremated bodies (15 total) this study utilized wooden caskets and a multi-chambered retort.³

The purpose of this research was to identify a predictable progression of thermal destruction characteristics to human bodies cremated in a commercial environment, thereby establishing a baseline for analysis of forensic significance in unexplained deaths. Further, statistical analyses were undertaken to determine if burn progression is correlated with cadaver weight, sex, or the position of the body in the fire.

Between September 2010 and March 2011, nine cremations were analyzed at a commercial crematorium in Pennsylvania as part of NIJ award No. 2008-DN-BX-K131. Through videography, still photography, and visual observation, data were collected and compiled. Utilizing these data, eight thermal modifications occurring with the highest frequency were identified for analysis. These events included pugilistic posture of the hands, disarticulation of the hands, disarticulation of the thumb, pugilistic posture of the lower limbs, femur fractures, rib end fraying, exposure and destruction of cranial bone, and disarticulation of the lower limbs. Body position and supine/prone positioning were examined. Ranked data were then evaluated using Spearman's Rank Correlation between each case to look for a quantifiable pattern in burn progression.

Despite a small sample size, initial analyses indicated

that expected thermal characteristics were predictable due to soft tissue shielding and body position. Each was readily identifiable and predictable in progression. Interestingly, there did not appear to be a high correlation between individuals of the same sex, nor did body weight or age have a strong influence on the progression of the burn. However, there was a correlation between cases with similar body positions, indicating that the positioning of the body plays a role in burn progression. Generally, pugilistic posture of the limbs occurred early in the cremation process with restrictions of the limbs occurring in those bodies positioned face down. Also, fraying of rib ends depended on body position. The sternal-end frayed in the supine position, while the vertebral-end frayed in the prone position.

Predictable burn progression was supported by statistical analysis when events were ranked by order of occurrence, resulting in similar burn patterns across multiple cases in similar settings. Body position appears to play a role when variability in expected burn patterns is exhibited. Despite predictable patterns based on body position and tissue protection features, further research in non-commercial, natural settings would contribute to anthropological predictability of bone destruction in fires. Understanding the expected patterns of thermal destruction is important to accurately distinguish between thermal damage and non-thermal peri-mortem bone trauma in forensic cases.

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Cremation, Thermal Destruction, Burn Progression

H136 Transformation by Fire: Posthumous Treatment and Biological Profile Reconstruction of Cremated Human Remains

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After attending this presentation, attendees will have a better understanding of: (1) variability in posthumous treatment and thermal alteration of highly fragmented and burned human bodies; and, (2) methods and protocols inspired by the study of modern and ancient cremations for the purpose of reconstructing biological profiles and postmortem treatments.

This presentation will impact the forensic science community by demonstrating the importance of an integrated approach to analyzing highly fragmented and burned human remains to obtain a positive identification, reconstruct postmortem events of legal importance, and conduct a successful deposition.

Anthropological analysis of highly fragmented and burned human remains is of great importance. Remains in these conditions are often produced in cases of mass fatalities, such as terrorist activities, and airplane crashes, among others. Individual cases of highly burned human remains involving accidental and/

or intentional fires also occasionally arrive at Medical Examiner Offices, requiring anthropological consideration. The type and degree of thermal alterations can be unique to particular cases but present some commonalities depending on the duration and exposure of the body to the fire. Nonetheless, on most occasions these types of cases are fairly complex due to the high degree of fragmentation and destruction of the body. Using an approach integrating study of the recovery scene, reconstruction of biological profile, and examination of thermal alteration and body manipulation can advance reconstruction of events, identification processes, and successful deposition. This study is informed by analytical methods developed through work with both modern and ancient cases of cremated bodies. This presentation, we illustrate this approach with two case studies from the Sonoran Desert of southern Arizona that present somewhat similar postmortem treatments. The first step of the analysis was a detailed skeletal inventory of each individual. These analyses allow for interpretations of body completeness and the minimum number of individuals represented in each deposit. Following this inventory a biological profile was produced in which age-at-death, sex, pathological conditions, degenerative joint disease, and trauma were assessed. Thermal alterations and posthumous treatment of the bodies were documented in detail. In this research, bone color, bone weight, type and degree of fractures cause by fire, maximum length (mm), average length (mm), and quantification of bone fragments $\geq 2\text{mm}$ in length for the cranial, dental, axial, upper appendicular, and lower appendicular anatomical regions were recorded. Analysis of the case deposits included a detailed reconstruction of the disposal area to distinguish between potential post-depositional disturbances, such as natural processes, soil condition (e.g., pH), and bioturbation, from intentional practices. Case A presents the analysis of two victims. These two victims were murdered and their bodies were thoroughly burned for several days in four different trash pits. Osteological analysis revealed the presence of approximately 3,210 countable, and many smaller, burned human bone fragments of different sizes from two adult individuals. The remains were differentially exposed to fire and highly fragmented. Morphological landmarks and differential thermal alterations on the human remains allowed for refitting of several bone elements, evaluation of differential exposure of the remains in the fire, and to reconstructing posthumous treatment of the individuals. Biological profiles allowed circumstantial identification of the two victims, and ultimately DNA analysis comparisons allowed that one individual could be positively identified. The case was successfully presented in court and a sentence was delivered to the defendant. Case B is an individual from the Prehispanic Hohokam culture (Preclassic Period, A.D. 700-1150), also from the Sonoran Desert of southern Arizona. In this case, the remains were highly burned and fragmented in a cremation funeral ritual and later the body of the deceased was divided and placed into at least two separate pits in the ground. Although this Hohokam individual was burned as part of funeral customs, the body presented similar thermal alterations, degree of fragmentation, and posthumous treatment as the individuals in Case A. Using the treatment of both modern and ancient cremated remains as case studies, this presentation seeks to demonstrate the potential advantages in the integration of forensic and bioarchaeological investigations to contemporary death investigation.

Cremation, Postmortem Treatments, Biological Profile

H137 Relocation of Remains: Scavenger Scatter Patterns in Central Oklahoma

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After attending this presentation, attendees will gain a better understanding of how scavengers disperse remains and the predictable patterns related to those behaviors, as well as key indicators of directionality of dispersal and exposure time, which will aid them in more successful search for and recovery of human remains which are left exposed at the surface level.

This presentation will impact the forensic science community by improving search and recovery techniques, increasing percentage of remains recovered, increasing identifications of remains, closing cases, and bringing closure to families and the community.

There are many instances annually where human remains are left exposed, due to a variety of situations deriving from accidents, suicide, and homicide. In these instances, the likelihood of scavenging of the remains is high. Scavenging results in the scatter of remains from the original deposition site, and in some instances this can cover a vast area, depending on species, terrain, and season, adding difficulty to search efforts. Patterns in this dispersal have been noted in few case studies as well as experimental cases. This study hypothesizes that this dispersal is highly patterned and predictable based on a series of multi-seasonal experiments where remains were constantly monitored and tracked in an accurately measurable way.

This study utilized wildlife radio transmitters in a novel and unique way, implanting them inside medullary cavities of long bones, attaching them to the surface of the pelvis, and implanting them within the crania of domestic pig carcasses which were left exposed. These skeletal elements were then able to be tracked over time, revealing the direction, movement, and final resting places of those skeletal elements throughout the decomposition and scavenging process. This allowed for the first time for typically lost skeletal elements to be followed and mapped from deposition day throughout the entire processes of decomposition and scavenger consumption. From the data collected, a pattern of use of the area by wildlife and subsequently pattern of maximum dispersal was observed.

This experiment was repeated several times over several seasons making the information in this presentation highly scientifically valid and vastly useful to the forensic science community in areas with similar ecologies and wildlife. The data collected over the four sets of experiments and the implications of that data and application in the field will be presented.

Scavenging, Skeletal Remains, Recovery Technique

H138 The Effect of Altitude on Decomposition: A Validation Study of the Megyesi Method

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The goal of this presentation is to explore the effect of altitude on decomposition. After attending this presentation, attendees will gain an understanding of the Megyesi method's lack of utility in Colorado's Rocky Mountain region.

This presentation will impact the forensic science community by demonstrating decomposition patterns between high- and low-altitude sites, providing data regarding scavenger

behavior, and establishing the regionally specific validity of the Megyesi method.

The Rocky Mountain region is a geographically distinct area characterized by ecozone convergence. Arid canyons, semi-arid grass plains, alluvial valleys, lava fields, volcanic plateaus, woodland hills, forested mountains, glaciated peaks, and a variety of aquatic habitats situated at elevations ranging from 1,010 to 4,400 meters result in closely oriented, dynamic micro-climatic zones. Complex interactions between human remains and the greater ambient environment in which they exist make every death scene distinctive. But, the laws governing the decomposition of human remains can be generally quantified, making the interaction with the environment the greatest variable in an individual case. Decomposition studies, while numerous, have largely been conducted in the southeastern and northwestern United States, and, therefore, the results are limited by geographic circumscription. There is currently a lack of published information on the effects of altitude on human decomposition. The Metropolitan State University of Denver Human Identification Laboratory (MSU Denver-HIL) has, therefore, initiated ongoing inquiries into regionally specific patterns of decomposition and taphonomic progression.

Utilizing four subjects (*Sus scrofa*), a concurrent study of decomposition rates among and between a high-altitude site (2800m) located in Gilpin County and a low-altitude site (1,200m) located in Keenesburg was conducted in July 2012. Over the course of 30 days, two observers participated in the daily completion of scopic-scoring worksheets for each of the subjects. Two observers were used in order to allow for the assessment of inter-observer error. Scavenger activity was documented at each site using motion-activated infrared game cameras, and data pertaining to arthropod progression was collected. Daily temperatures were obtained utilizing portable digital data loggers, and off-site through data provided by the local weather station most closely associated with each site. Both sources were used to calculate actual Accumulated Degree Days (ADD); only marginal deviation was observed between site-specific and site-relative temperatures. The Megyesi *et al.* regression equation was utilized to estimate ADD and the results were compared to the actual ADD using a two-tailed student's *t*-test.¹ The same statistical analysis was applied to daily total body scores and average daily temperatures between both the high- and low-altitude sites.

Preliminary results suggest that the Megyesi method is not effective for accurately calculating Postmortem Interval (PMI) at either the high-altitude or the low-altitude site, suggesting that an unaccounted for variable is driving decomposition rates within this geographically distinct region of Colorado. The cadavers at both sites reached a plateau in the decomposition process within one to three weeks resulting in dermal tanning at the high-altitude site and complete mummification at the low-altitude site. The difference between average total body scores between the two sites was not statistically significant implying altitude alone is not a significant factor affecting decomposition. The difference in average temperature between the two sites was statistically significant, and yet, there was not a significant difference between the total body scores suggesting temperature alone is not a significant factor.

Scavenger activity also diverged greatly from expected outcomes. Specifically, scavenger activity was significantly reduced at the high-altitude site and predominantly involved avian and rodent activity. While large scavengers such as black bears (*Ursus americanus*) were observed on numerous occasions, they consistently opted to exploit the arthropod larvae colonizing the remains as opposed to the remains themselves, suggesting that the cadavers were not a preferred food source. At the low-altitude site, scavenger activity was non-existent throughout the study period and mummification occurred rapidly. While adult coleopterans were observed at the site, grasshopper larvae were the primary colonizers of the remains. The atypical activity observed at both

sites suggests that numerous additional studies, including those involving dew point, humidity, aridity, and the agency of microbes as competitive scavengers, are necessary in order to understand the complex interaction between local agents affecting decomposition.

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Decomposition, Altitude, Megyesi Method

H139 Postmortem Interval Estimation Using ATR-FTIR and Raman Spectroscopy

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After attending this presentation, attendees will understand some of the techniques currently used to estimate the Postmortem Interval (PMI), a new method of estimating PMI in advanced stages of decomposition with spectroscopy, and the prospects of this research for the future.

This presentation will impact the forensic science community by providing an objective, reliable, and quick method of determining the time since death, which aids in identifying human remains, and resolving other aspects surrounding the death of the individual.

Estimating PMI is accomplished in a variety of ways, including scoring visual characteristics of decomposition, assessing characteristics of the environment in which a corpse is discovered, and analyzing the chemical components of soft tissues. As a body decomposes, the methods available to estimate PMI dwindle and become less accurate. Vibrational spectroscopy offers a simple, objective alternative to estimate PMI throughout the course of decomposition.

Vibrational spectroscopy includes a group of analytical techniques which identify the molecular components of a substance by passing light through the sample and measuring the energy change in the vibrations of chemical bonds as the bonds are distorted by the light energy. Raman and Attenuated Total Reflectance Fourier Transform Infrared (ATR-FTIR) spectroscopy are rapid, non-destructive techniques that yield results in minutes with minimal sample preparation.¹ Previous studies have successfully utilized ATR-FTIR spectroscopy to determine PMI from the kidney tissue of rats.² These results suggest that spectroscopic techniques may be able to estimate PMI of fluid samples after the body has reached an advanced state of decomposition and soft tissues no longer retain their integrity.

For this study, twelve domestic piglets, *Sus scrofa*, with a mean weight of 6.74kg were placed in plastic trays and allowed to decompose. Six pigs were placed in the shade and exposed to the environment; six pigs were placed indoors with an electric radiator ensuring an elevated temperature. In both experimental set-ups, insects had access to the carcasses, although access was delayed by approximately 15 ADD in the indoor group. Data loggers in the trays next to each pig recorded the temperature every hour. Fluid and tissue were sampled approximately every 20 Accumulated Degree Days (ADD) for 21 days, at which time the bodies were mostly skeletonized (mean Total Body Score = 30.25). All samples were frozen and then thawed prior to spectroscopic analysis. Samples were analyzed with both Raman and ATR-FTIR spectroscopy from 4000 – 400nm. Fluid was placed directly onto a diamond crystal and allowed to dry prior to ATR-FTIR analysis. Fluid for Raman analysis was placed onto CaF slides and allowed to dry before analysis with a 532nm laser.

The spectra obtained from the ATR-FTIR and Raman analyses were subjected to principle component analysis. Preliminary results indicate that the first three components can be used to classify samples based on ADD up to skeletonization where the fluid was available for collection in the plastic trays. The first three components explain up to 94% of the variance. This provides promising continuum of change from which a method to pinpoint PMI can be derived from the principle components.

Therefore, spectroscopy is a valuable tool in estimating the PMI of corpses from fresh to skeletonized in a variety of environmental conditions with available fluid. The results are objective and are available in minutes. As this technology advances and becomes more portable, investigators may be able to receive an estimate of the PMI while in the field at the initial visit, which may speed investigations and require fewer resources to solve each case.

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Postmortem Interval, Raman Spectroscopy, FTIR Spectroscopy

H140 A “Realistic” Study of Sharp Force Trauma Recognition in Burned Remains: The Forensic Implications

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After attending this presentation, attendees will gain an understanding of the effects of fire on trauma in bone and of the methodologies utilized in corresponding experimental forensic anthropological research.

This presentation will impact the forensic science community by demonstrating what the effects of fire modification are on Sharp Force Trauma (SFT) in bone, specifically the thoracic region of fully fleshed and clothed remains burned in a realistic fire scenario. This presentation will also highlight that caution should be used when designing forensic experimental studies and assurances should be made that experimental conditions reflect forensic reality.

Research into fire modification of bone is necessary in forensic investigations, as clandestine fires can be used to cover up a homicide by attempting to destroy evidence of trauma and contact to the perpetrator or identity of the victim.^{1,2} Previous research studies have utilized (partly) defleshed bones and fire substitutes such as furnaces and gas cookers in their research into burned bone and/or SFT.³⁻⁵ These conditions don't reflect actual crime scenarios; therefore, this study proposed to mimic forensic reality by using whole, clothed remains, stabbed in the thorax, and burned in a real fire. Research was conducted with help of the West Midlands Fire Service Research and Investigation Unit. This study presents the findings of a study that intended to determine the effect of fire modification on SFT, expecting warping, color change, and fracturing of the bone and trauma location.

To create a realistic experimental design, freshly slaughtered fleshed and clothed *sus scrofa* were stabbed using two

knives (a serrated and a non-serrated kitchen knife) in the thoracic region, reflecting the most common homicide type in England and Wales.⁶⁻⁸ Two fire scenarios were used: an accelerated pyre and a body doused in white spirits and left on the ground, to represent a body dump. The fires were left to burn for 2 hours. The West Midlands fire service extinguished the fires as they would any other fire of this kind. The remains were removed from the fire, bagged, and taken to the lab where the ribs were excised and cleaned. Following cleaning, analysis of the ribs was possible.

Unexpectedly, there was little to no fire modification of the bones, let alone of the trauma. Sample A, burned on the pyre, exhibited severe burning to level 3 of the Crow-Glassman scale; the forelimbs, cranium and vertebrae were severely burned.⁹ Despite this extensive burned exterior, the ribs were safely encased in the surrounding flesh, with only some charring at the stab sites. Sample B showed only minor charring of the skin, although fat had escaped from the stab wounds and rendered down; the remains had failed to “burn.” There was no fire modification of the ribs of sample B.

The results of this experiment were surprising given the findings of previous research in this field and the similar, albeit more realistic and larger scale experimental approach taken in this study. There was no notable fire modification of the SFT present in the ribs. These findings raise questions regarding the validity of conducting forensic research that does not sufficiently incorporate forensic reality of an actual crime scenario into the experimental design, and the implications of these findings for forensic science research will be discussed in the presentation.

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Forensic Research, Sharp Force Trauma, Fire

H141 Experimental Wood Chipper Reduction: Trauma and Distribution Patterns

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After attending this presentation, attendees will understand the expected trauma and distribution patterns of hard and soft tissues associated with the reduction of human remains

using a small, commercial, disc-type wood chipper applied to a porcine model.

This presentation will impact the forensic science community by providing basic guidelines for the skeletal and integumentary trauma patterns of tissues subjected to wood chipper trauma. It will provide expected distribution patterns of remains that will help crime scene investigators, law enforcement, and forensic anthropologists recover the maximum amount of hard and soft tissues possible. This study also outlines the possible ease of human-enacted and natural methods of concealment. In addition, this presentation illustrates the different trauma patterns expected for the axial versus appendicular skeleton placed into a wood chipper, including size ranges. The information included in this presentation is important for the search, recovery, analysis, and contextual information associated with traumatic injury wood chipper cases.

Previous studies have focused on fragment size and the differentiation between chipping and shredding mechanisms and the macroscopic trauma characteristics applied to long bones by home-model wood chippers.^{1,2} A single 300-pound *Sus scrofa domestica* carcass was acquired from a local farm and dismembered at the Forensic Anthropology Research Facility at Texas State University. The limbs were dismembered at the joints to avoid creation of confounding trauma characteristics. Ribs were disarticulated from the vertebrae near the rib heads with a pair of loppers. Only lateral and anterior rib fragments were sampled to avoid confounding variables. Following dismemberment and evisceration, the appendicular and axial portions were placed into the machine separately with the chute moved in between chipping of the two groups. Bones were placed into the machine proximal portions first, mimicking a person entering head-first into the wood chipper. The resulting distributions were mapped and measured separately with the appendicular skeleton as distribution "a" and the axial portion as distribution "b." The distributions were kept separate throughout the warm water maceration process. Forty macerated sample fragments from each distribution were examined for macroscopic and microscopic characteristics of sharp and blunt force trauma. Observed trauma characteristics included kerf marks, longitudinal and transverse fractures, through-cuts, spurs, and notches.

In this study, there were observable differences in the frequency of both microscopic and macroscopic traumatic characteristics of the axial versus appendicular skeletal material. Fragment size ranged from a large 84mm long rib fragment to skeletal debris the size of sand. Many fragments were macroscopically identifiable by skeletal element as well as human versus non-human origins. In addition to skeletal fragments, large swatches of skin as large as 260mm by 170mm were also recovered from the distribution area.

While the use of wood chippers as tools for human remains reduction is uncommon, these machines have been used in the past to conceal homicides. Some cases are still under investigation today. The expansion of knowledge regarding wood chipper reduction as reported in this presentation could expedite the process of remains recovery and analysis by guiding investigators toward a more accurate search area and providing expectations as to fragment size and trauma patterns. This study will present an account of experimental wood chipper trauma and the results regarding expected fragment size in axial versus appendicular bone and skin, the distribution pattern of remains, and microscopic as well as macroscopic skeletal trauma patterns.

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Skeletal Trauma, Wood Chipper, Bone Fragments

H142 Effective Use of Metadata in the Identification of Skeletal Remains With Weak DNA Kinship-Index Results

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After attending this presentation, attendees will have increased understanding of the utility of metadata in the personal identification process at a large urban medical examiner's office and the types of metadata that may be used effectively to support identification when DNA results are inconclusive. Retention of this information will be enhanced by the presentation of a complicated case series in which metadata were used effectively to assist in identification of skeletal remains.

This presentation will impact the forensic science community by increasing the potential for successful identification of deceased persons through the demonstration of innovative uses of metadata in personal identification of skeletal remains with degraded bone and limited family-reference sample availability.

In general, metadata is a term for information that describes other data. In the context of forensic personal identification, metadata are forms of evidence that may aid in interpretation of molecular evidence, i.e., DNA profile comparison results. Metadata are often provided to medical examiners by a consultant or staff forensic anthropologist in the form of the anthropologic profile, a comprehensive demographic description of an unidentified deceased individual in life constructed through observation and interpretation of metric and non-metric evidence preserved in the skeleton. Characteristics beyond age, ancestry, stature, and sex of the remains are assessed, including trauma, pathological conditions, anomalies, and stress markers. The use of DNA profile comparison for identification of skeletal remains has become standard procedure in many United States medical examiner offices over the last 20 years. The anthropologic profile reduces the pool of possible identities and may be used to simplify the search for potential family members who will donate family reference samples for DNA analysis. Once the DNA comparison is completed, a Kinship Index (KI) is reported by the laboratory to aid in the identification decision. When the KI is inconclusive, the metadata may support or cast doubt on the presumptive identity of the skeletal remains.

Harris County Institute of Forensic Sciences located in Houston, Texas, houses the Harris County Medical Examiner's Office (HCIFS). An unidentified skeletal remains case of a teenaged male killed during the "Houston Mass Murders" of 1973 (778) was identified in 2010 by HCIFS through DNA comparison with a living sister's profile. The sister's reference sample was collected following recognition of a similar dental non-metric trait pattern in photos of another deceased sibling. Although no dental records were available, photos clearly showed that both young men had both shovel-shaped incisors and pronounced bilateral Carabelli's cusps. The identification of 778 illuminated the 1973 misidentification of a different skeletal case (733). Investigation of the misidentification required exhumation of two teenaged male decedents who had been buried in the same casket (733 and 713). One of the decedents, 733, had been previously misidentified with the same identity confirmed through DNA for 778. The exhumed decedents were skeletal and the remains had been subjected to ebbing and flowing groundwater, degrading the DNA. The complete mitochondrial and partial nuclear DNA profiles obtained from 733 were compared with the profiles from a living sister but the KI was inconclusive. No other family references were available.

The anthropologic profile was highly consistent with the medical records and descriptions of the unidentified boy. Additionally, the decedent had undergone multiple Electroconvulsive Therapy (ECT) treatments with a hard rubber mouth guard. The anterior dentition was marked by patterned wear and fractures inconsistent with his age. The wear pattern and fracture locations were found to be consistent in comparison with that of another identified teenaged male from the time period (750) who received multiple ECT treatments. Based on metadata support of the KI, the decedent was positively identified. Although these cases are 40 years old, ECT is currently in widespread use as a treatment for depression. Recognition of ECT-patterned wear and fractures may also provide supporting metadata in current casework.

Personal Identification, Electroconvulsive Therapy, Dental Non-Metric Traits



11 The Use of Biomarkers in Clinical Settings: Ethical Reflections

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After attending this presentation, attendees will be able to recognize characteristics in the use of biomarkers in neurodegenerative diseases.

This presentation will impact the forensic science community by demonstrating ethical reflections on the dichotomy between the principle of autonomy and the principle of beneficence.

Until recently, the notion of having blood markers of Alzheimer's Disease (AD) available for clinical use was more likely to be seen in an episode of a TV medical drama or a science fiction movie, than in a peer-reviewed scientific journal.

"Biomarkers" are quantifiable biological substances, characteristics, or images which provide an indication of the biological state of an organism. They are objectively measured and evaluated as indicators of normal biological processes, pathogenic processes, or pharmacological responses to therapeutic interventions. The search for biomarkers has suffered from serious obstacles, including, but not limited to, inadequate diagnostic accuracy and inabilities in the replication of findings across samples between laboratories or even within individual laboratories.

Biomarkers offer a measure of biological functions that can help to unravel a mechanism or biological pathway. They can serve as predictors of the disease's future course. They can also provide indications of both potential effectiveness and potential hazards associated with therapeutic interventions.^{1,2} In neurodegenerative diseases, biomarkers represent promising approaches to staging diseases, in order to identify clinical subgroups. Current clinical research focuses on the use of biomarkers to improve on the early detection of diseases as well as pre-symptomatic detection of neuronal dysfunction.³ Over the years, the diagnosis of AD as well as other neurodegenerative diseases has undergone significant changes in relationship with scientific advances in the fields of biomolecular engineering, neuropathology, and neuroimaging. Especially in the last 25 years, diagnostic criteria have undergone significant advances in their classification schemes. In particular, many neurodegenerative diseases present themselves many years before the onset of clinically significant signs and symptoms. Especially in the context of AD, it is known that the AD pathophysiological process likely starts years before cognitive changes and decades before the onset of clinical dementia. Biomarkers of AD are available in some medical centers, but they are not routinely utilized in clinical practice.

The ethical debate related to this topic stems from the

dichotomy between the principle of autonomy (respecting a patient's right to self-determination, including the right to know) and the principle of beneficence (producing benefits or favorable outcomes for a patient). To ensure the possibility of using biomarkers in the clinical setting consistent with both of these bioethical principles, this study has created formats for information sheets and informed consent. Consideration of the possible value of biomarkers in early diagnosis of, as well as monitoring tools for, neurodegenerative diseases is encouraged. This study's formats also emphasize that some biomarkers' identification (e.g., via blood or cerebrospinal fluid) are not necessarily definitive diagnoses and that, at present, their validity is not entirely certain.

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Biomarkers, Bioethical Principles, Informed Consent

12 Genetic Study of DNA Polymorphisms in Androgen Receptor, Serotonin Transporter, and Monoamine-Oxidase Genes in an Inmate Sample

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After attending this presentation, attendees will understand how analyzing specific genetic polymorphisms located in genes associated with aggressive behavior can lead to significant differences between an inmate sample and controls.

This presentation will impact the forensic science community by revealing a possible mechanism that would explain the predisposition for aggressive behavior which may provide valuable information for criminal behavior analysts and profilers. Studies show that self-reported questionnaires are the most commonly utilized technique for analyzing the level of aggression in an individual. A major disadvantage to this technique is the uncertainty that the reported values reflect the true behavior of the individual. Therefore, research focused on DNA profiling to determine an individual's predisposition for aggression may be useful to understand the mechanisms which regulate aggressive

behaviors.

Emotional responses are controlled by a complex system of neurotransmitters in the brain. Genetic polymorphisms located in the coding genes of neurotransmitter receptors, enzymes, and transporters have been shown to modulate the transcription of these proteins. The interaction and modulation of central neurotransmitters and related proteins (receptors, transporters, and metabolic enzymes) affect emotional behaviors such as aggression. Associations between aggressive behavior and specific polymorphisms on the Androgen Receptor (AR), the Monoamine Oxidase A (MAOA-VNTR), and the serotonin transporter (5-HTTLPR) genes have been previously reported. This research intends to compare the allele frequencies of repeat variants located in AR, MAOA, and 5-HTT genes between inmates and a control population of Texas. Buccal swabs were collected from male inmates incarcerated at a jail located in southern Texas (N=98) and from control male students at Sam Houston State University (N=93). All samples were extracted using organic extraction with ethanol precipitation, quantified, amplified, and then analyzed by capillary electrophoresis with fluorescent detection. For 5-HTT, departures from the Hardy-Weinberg equilibrium were detected in the inmate sample ($p < 0.01$) but not in the control group. Significant differences were observed in allele frequencies of MAOA-VNTR ($p < 0.05$) and AR ($p < 0.01$) but not in 5-HTT, when both samples were compared. MAOA alleles 2 and 3 showed higher frequency in the inmate sample. Research has shown that alleles 3.5 or 4 are transcribed more efficiently, whereas alleles 2, 3, or 5 show lower levels of transcription. Allele 4 has been classified as a high-activity MAOA allele, while alleles 2 and 3 are known as low-activity MAOA alleles. The low-activity MAOA has been associated in past studies with increased levels of antisocial and criminal behavior. Higher frequency of shorter AR alleles (<23 repeats) was also detected in the inmate group. Previous studies have shown that shorter Cytosine-Adenine-Guanine (CAG) repeats are associated with impulsive-disinhibited personality traits and increased verbal aggression. Although no significant differences were detected in 5-HTT, the very long allele was observed only in the inmate sample. The results obtained for the AR and MAOA polymorphisms support the hypothesis that the inmate population would display a higher frequency of short repeats which, in turn, is associated with aggressive behavior.

However, contrary to the hypothesis, no differences were observed for the 5-HTT polymorphism. This discrepancy may be attributed to the limited size of the sample. Overall, the results support current studies in human and animal models; however, much research is still required to understand the complex mechanisms which regulate aggressive behaviors.

Monoamine-Oxidase, Serotonin Transporter, Androgen Receptor

13 47, XYY Karyotype and Deviance: A Case Report

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After attending this presentation, attendees will be able to discuss the influence of genetics on criminal behavior and will understand how to approach a case of psychiatric evaluation on a subject affected by a chromosomal abnormality.

This presentation will impact the forensic science community by demonstrating how clinical evaluation can promote

accurate and complete knowledge of an individual.

The case report concerns a young Italian man afflicted by Borderline Personality Disorder and Jacobs' Syndrome, which consists of a 47,XYY karyotype. The additional Y-chromosome derives from paternal meiosis II non-disjunction or postzygotic mitotic non-disjunction.

The prevalence of XYY Sex Chromosome Abnormalities (SCAs) in newborns is as high as 1:1000, but they are often unidentified because they are not necessarily associated with gross physical or cognitive impairments, and may never come to the attention of medical personnel. The only common and obvious features are high stature (greater than six feet) and a strong build.

The young man in this case report frequently exhibited violent behavior toward his relatives, and the court asked for an evaluation of his mental condition. Forensic psychiatrists expressed their opinions regarding his danger to society and criminal liability by analyzing the influences on his behavior from both his mental and genetic conditions. During the 1960s and the 1970s, some studies postulated that Jacobs' Syndrome can lead to aggressive behavior, but the statistical certainty of this association was questioned. Some experts hypothesized that these subjects' increased tendencies to commit crimes is not due to underlying aggression, but rather to associated intellectual deficits, since the presence of an additional Y-chromosome is related to cognitive disorders and deficits in intelligence. Other authorities hypothesized that these men are more easily recognized as offenders because of physical characteristics (e.g., high stature and stocky build), making them more identifiable.

More recent developments in genetics and neuroimaging have led to new publications on the relationship between the presence of an extra Y-chromosome and social function. According to the results of these studies, there is no significant evidence that people affected by Jacobs' Syndrome are necessarily antisocial or deviant. Such behaviors are indeed thought to have multifactorial etiologies, in which the genetic component may play some role, but should not be considered the sole cause.

Even today, there are differing opinions about the relevance of Jacobs' Syndrome in the forensic psychiatric assessment of societal danger and criminal liability, hence the importance of clinical evaluations and findings to promote an accurate and complete knowledge of individuals. This presentation concludes by reporting the accused's statements, revealing his personal feelings about the role his karyotype has played in influencing his criminal behaviour.

Jacobs' Syndrome, Genetics, Deviance

14 Neurogenetic Basis of Criminal Behaviors in Klinefelter Syndrome: A Case of Uxoricide-Suicide and a Review of the Literature

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After attending this presentation, attendees will be able to better understand cases of Klinefelter Syndrome.

This presentation will impact the forensic science

community by demonstrating a correlation between chromosomal abnormalities and criminal behaviors in Klinefelter Syndrome.

Klinefelter Syndrome ("KS," XXY) is the most common sex chromosome abnormality in men. It occurs with a frequency of 1:500 to 1:1000 live male births. The prevalence of Klinefelter Syndrome is around 150 per 100,000 males or one in every 660 males. This chromosomal disorder is characterized by the presence of one extra X chromosome (or more) in a phenotypic male. The most prevalent type of karyotype is 47, XXY.¹ The physical phenotype is characterized by tall stature, small testicles, cryptorchidism, hypogonadism with low levels of testosterone, infertility, gynecomastia, broad hips, narrow shoulders, and sparse body hair.² The abnormalities of the sex chromosome also lead to alterations of brain development. This process may be a genetic basis of the psychiatric disorders described in these subjects. The cognitive phenotype of KS is characterized by language learning problems and mental retardation.³ In these subjects, there is an increased risk of psychiatric disorders, such as symptoms associated with schizophrenia, which are likely to be related with the over-expression of X-linked genes. Patients with KS also show an increased risk of criminal behavior and, in a significant proportion of cases, psychosocial problems have been reported.⁴ The most common offenses reported to be related to KS are sexual abuse, arson, burglary, homicide, and drug-related crimes. KS is often under-diagnosed. The goal of this study is to verify the hypothesis of a correlation between chromosomal abnormalities and criminal behaviors through the analysis of a forensic case of uxoricide-suicide.

Case Report: This study analyzed the case of an older man found dead in his cottage with a gun in his right hand. Judicial inspection revealed a suicidal, single gunshot injury. Social background was reconstructed by psychological autopsy. Data analysis highlighted a personal story of uxoricide, and his life history was surveyed. The investigation demonstrated a medical history of infertility. An analysis of his pending criminal proceedings revealed an incident of uxoricide about 50 years prior (1962). The court's archives were searched for statements made by the man (then-husband) when interrogated. The records revealed that the 25-year-old woman was killed by her husband using a single gunshot injury. She was transported to a nearby hospital, and the doctors found "internal bleeding from gunshot wound penetrating into the abdominal cavity." In the intervening time, her husband vanished but was later arrested.

Autopsy investigation of the man in this case was carried out, and the typical phenotype associated with KS was found. The postmortem clinical diagnosis was confirmed through the histopathological and genetic analysis of his karyotype.

Conclusions: The retrospective analysis of the case showed a correlation between this syndrome, psychiatric disorder, and criminal activity.

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Klinefelter Syndrome, Karyotype, Psychiatric Disorders

15 Measuring the Mind of a War Criminal: Behind the Scenes on an Assignment for the United Nations International Court of Justice in the Hague

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After attending this presentation, attendees will have a greater insight into the process and practice of consulting for the United Nations International Court of Justice, including dealing with cross-cultural issues, language barriers, bias-reduction through culture-free test selection, and communication of findings that address international criminal-legal standards.

This presentation will impact the forensic science community by sharing insights into the process of forensic neuropsychological consultation on an international level.

This paper presents a first-hand account of an assignment from the United Nations International Court of Justice in the Hague to evaluate the competency of a Bosnian war criminal to participate in an appeal of his case. The history and role of the International Criminal Tribunal for the former Yugoslavia (ICTY) in this context will also be described.

An orientation to the 1995 "ethnic cleansing" and genocide in the town of Srebrenica in Bosnia and Herzegovina will be provided. This mass murder involved the killing of more than 8,000 Bosnian Muslim men and boys by members of the Army of the Serbian Republic and the mass expulsion of another 25,000 to 30,000 Bosnian Muslim civilians. In addition, according to the ICTY indictment, victims endured: unlawful confinement, murder, rape, sexual assault, torture, beating, robbery, and inhumane treatment; the targeting of political leaders, intellectuals, and professionals; the unlawful deportation and transfer of civilians; the unlawful shelling of civilians; the unlawful appropriation and plunder of real and personal property; the destruction of homes and businesses; and, the destruction of places of worship.

The procedural history of the instant case will be discussed, with a focus on the international criminal-legal mental health standards for competency to pursue appeals in this context. In addition, the issues leading to the referral for a forensic neuropsychological examination in the case will be discussed. This will include a review of the relevant *a priori* medical and forensic psychiatric findings in the case, and the issues which arose from there, driving the need for this assignment.

The logistics of arranging travel, accommodations, security, the examination location, and coordination with international attorneys and United Nation (U.N.) representatives will be presented, including what happens when one's luggage gets lost in Serbia. Insights with regard to maintaining objectivity and professional neutrality in such emotionally-charged circumstances will also be shared.

Specific challenges encountered during the assignment will be explored, including: (1) cross-cultural issues in conducting the forensic neuropsychological examination; (2) addressing reliability and validity concerns through selection of culture-free test instruments and measures; (3) issues of literal language translation; and, (4) interdisciplinary consultation with experts from other countries.

Findings from the examination will then be summarized.

This will include personal observations and a phenomenological account of the examination itself, as well as a discussion of how the results from the neuropsychological test battery were directly related to the issues of trial competency before the court. Finally, the process of report writing and communication of findings to the Appeals Chamber of the International Criminal Tribunal for the former Yugoslavia will be discussed, together with the Appeals Chamber's decision and the outcome of the case.

Forensic Neuropsychology, Bosnian Genocide, International Court of Justice

16 True Faces of Terrorists

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After attending this presentation, attendees will: (1) gain understanding of terrorism against the world, its allied governments, and global interests and the "3 I's" that distinguish terrorism from crime and other criminal actions; (2) gain an enhanced knowledge and understanding of the Middle East (psychological and sociological explanations of terrorism); (3) develop basic knowledge of clinical and social psychology as frameworks for analyzing terrorism and terrorists; (4) be able to analyze the causes of terrorism, the motivations of terrorists, and known international terrorist networks; (5) develop an understanding of and sensitivity to the social-political conditions that spawn terrorist organizations; (6) be able to explore the psychological processes that create a terrorist; (7) be able to examine the psychological impact of terrorist activities in prison; (8) be able to appreciate the need for further understanding of terrorist events; (9) develop unbiased and analytical thinking capabilities of terrorist's psychology; (10) gain an increased awareness of cultural and multi-cultural issues through study of how social problems and social movements are related to, and affect, minority groups; and, (11) learn the four types of terrorists in terms of definition, assessment, and categorizing.

From direct interviews, this is a study of terrorist personalities and their involvement within the terrorist cell, including types, tactics, and trends on a worldwide scale as well as domestically. This presentation on terrorist psychology using a clinical and social psychology will impact the forensic science community by providing a framework for studying terrorist individuals and groups, terrorist origins, goals, dynamics, and ideologies. Work for this presentation involves examination of the structure and dynamics of terrorists in the Middle East (not to say that terrorism is only in that region of the globe).

Terrorist influences via the use of the media, religion, and politics will be presented. Participants will view different definitions and typologies of terrorism, from the eyes of those who are called terrorists, clandestine freedom fighters, perpetrators, extremists, and other titles.

The audience will view case studies of various terrorist individuals and groups to demonstrate abilities and capabilities to research a terrorist problem, profile, or scenario in-depth, and argue for suggested counterterrorism policy or negotiations strategy.

Questions to be addressed will include:

- What groups are active in this generation, and have they achieved their goals?
- What are some of the important cultural differences that should be considered by terrorists when attacking countries or "representatives" of such nations?
- Can there be some important cultural differences which should be considered when seeking to understand counter-terrorist

strategies?

- How do political and religious beliefs sometimes aid terrorists?
- Is the use of terrorist tactics ever justified?

There have been too many years of terror with differing views of the terrorists, with each sector of the globe interpreting the behavior similarly. However, there may be lessons learned in the analysis of conflict when people impose their own culturally-conditioned cognitions and emotions.

Today's "bad people" are on the internet reading behavioral analysis texts to learn how experts are trying to catch them. As a result, they set their standards at a different level to bypass the game plan of the "good guys."

Categories or classifications are psychopathic, ethno-geographic religious, ethno-geographic political, and retribitional terrorists. In the first, one works for the money and the power. The second and third have a shared purpose: to bring significant change to the world and to be rewarded. The fourth has with no pre-morbid characteristics of psychological disorders or disease, but has been directly associated with harm (loss of family, friends, and/or community and seeking revenge).

The ethno-geographic terrorists function with the "3 I's":

- Impress with their ideas or power on persons;
- Impose their beliefs and conditions on community; and
- Implement their system as control on the world.

When interviewing these characters face-to-face, the professional examiner recognizes the individual level of intellectual functioning, emotional stability, cognitive commitment, and behavioral obedience. This program goes beyond the Rome Memorandum.

This presentation will reveal the true words and conditions of people legally labeled as terrorists: why they joined clandestine groups, their commitment to the leader or to a cause (or both), and what might make them change directions toward non-violent extremist positions.

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Terrorist Psychology, Terrorist Cognition, Terrorist Behavior

17 (Un)Reliability in Forensic Mental Health Evaluations: Evaluator Variability and Adversarial Allegiance

Daniel C. Murrie, PhD, 1330 Amber Ridge Road, Charlottesville, VA 22901; and Marcus Boccaccini, PhD, SHSU Department of Psychology, Box 2447, Huntsville, TX 77341*

After attending this presentation, attendees will be familiar with evidence of unreliability in forensic mental health evaluations, as identified in field research and experimental research.

This presentation will impact the forensic science community by helping attendees better identify and minimize adversarial allegiance and other forms of bias in their work. Rarely has prior research explored bias in forensic mental

health evaluations and this presentation summarizes the first programmatic research on the topic, which carries implications for all forensic sciences.

Recently, the National Research Council warned that many popular forensic sciences may be unreliable or prone to error, and that forensic scientists are prone to bias because they lack independence from those requesting their services.¹ The NRC report did not specifically address the behavioral sciences, but their concerns are certainly generalizable to mental health experts and forensic mental health evaluations.

This presentation reviews a program of research that documents two sources of unreliability in forensic mental health evaluation. The first involves general evaluator differences, or idiosyncrasy. The second involves “adversarial allegiance,” the pull for experts to reach conclusions which support the party who retained them.

First, field studies strongly suggest evaluator differences, or idiosyncrasy, in forensic evaluations of trial competence, legal sanity, psychopathy, and sex offender risk. Second, related field studies strongly suggest “adversarial allegiance” in contexts where opposing evaluators assessed the same sexual offenders using the same risk assessment instruments. That is, experts on opposing sides tended to assign scores in a manner that supported the party who retained them.

These field studies raise an important question that can only be answered by a true experiment. Is apparent allegiance in courtroom trials due simply to attorneys choosing experts who have pre-existing attitudes which favor their side (selection effects)? Or do experts, once retained and promised payment by one side, tend to form opinions that favor that side (allegiance effects)?

A recent experiment deceived 100 forensic psychologists and psychiatrists to believe they were performing sex offender risk assessments for the prosecution or the defense. But, unbeknownst to them, all were scoring the same four offender case files, and all had been randomly assigned to believe they were working for the prosecution or defense. Just as in the field studies, evaluators who believed they were working for the prosecution assigned higher risk scores, and those who believed they were working for the defense assigned lower risk scores, even when applying the same instruments to the same offenders. Results provide strong evidence of adversarial allegiance. Follow-up surveys suggested that experts were aware of allegiance in general, but often had a “blind spot” regarding their own vulnerability to bias. This presentation summarizes this line of research, concluding with implications for all forensic science experts.

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1. National Research Council. *Strengthening Forensic Science in the United States: A Path Forward*. Washington, DC: The National Academies Press, 2009.

Bias, Adversarial Allegiance, Reliability

18 Delusions of the Devil or Malingered Psychosis? Cultural Considerations in Forensic Evaluations

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After attending this presentation, attendees will understand the importance of considering cultural context in forensic evaluations. Attendees will need to keep in mind the individual's cultural background when making psychiatric diagnosis, forensic formulations, and recommendations. Attendees will also have a better understanding of why ethnic background and religious beliefs

should be taken into consideration when diagnosing malingering.

This presentation will impact the forensic science community by elucidating the reasons why cultural context is important in forensic evaluations.

Forensic psychiatrists and psychologists are frequently asked to evaluate defendants from various cultural and ethnic backgrounds. Just as it is important in the clinical realm, good forensic evaluations need to appreciate the cultural context of the person evaluated. It is important to understand at least some of the cultural beliefs of the defendant in order to make psychiatric diagnoses, differentiate real psychiatric illness from malingering, assess the ability of the defendant to understand specific concepts, and generate formulations and forensic opinions. For example, for some cultures, it is more accepted to turn to religious leaders instead of mental health care providers when they are experiencing symptoms of depression, anxiety, or psychosis.

Sometimes, cultural norms influence the expression of symptoms. For example, if mental illness is not accepted in a culture, the patient may develop more somatic complaints of stress or depression. For instance, instead of acknowledging feelings of anxiety or depression, they may develop headaches, poor appetite, and insomnia. If a person is very religious and they become psychotic, they may believe they are having a negative or positive religious experience, such as hearing the voice of God or believing they are becoming possessed by the Devil. When assessing malingering in patients from different cultural backgrounds, it is important to keep cultural context in mind as well. Psychological testing is often used to assess malingering. However, it is important to understand how the testing is standardized and scored. Many of the tests are biased toward the dominant culture's knowledge and beliefs.

When updating the *Diagnostic and Statistical Manual of Mental Disorders* from IV to 5, a cultural issues subgroup was developed, “designed to improve the assessment of culture and context within psychiatric diagnosis.”¹ Aggarwal also discusses the importance of adapting the cultural formulation for clinical assessments in forensic psychiatry when performing forensic evaluations.² These articles will be discussed in this presentation. In addition, specific examples will be provided, illustrating the importance of keeping cultural context in mind when evaluating defendants for purposes such as competence to stand trial and “not guilty by reason of insanity.”

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Culture, Forensic Psychiatry, Malingering

19 Understanding the Mathematics and Economics of Malingering: Games in the Forensic Hospital

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After attending this presentation, attendees will become familiar with the understanding and application of game theory to the law, particularly in the realm of patients who malingering in forensic hospitals. Attendees will understand how environment of care, systemic forensic hospital factors, and treatment issues influence

the forensic hospital treatment “game” model. Finally, implications for individual management and hospital policy will be discussed.

This presentation will impact the forensic science community by supporting the development of a reductionist model of the forensic hospital malingering “game,” so that instrumental factors at play can be clearly and directly understood. Furthermore, the model will provide a discrete and reproducible method for the clinician and hospital administrator to identify and curtail malingering, allowing scarce forensic clinical services to be appropriately and optimally utilized.

Game theory can be formally defined as “the study of mathematical models of conflict and cooperation between intelligent, rational decision-makers.” The principles of game theory are generalizable to almost any social or interactive process and uses range from classical applications in economics, political science, and psychology, to more contemporary applications such as evolutionary biology and philosophy. In the language of game theory, a game refers to any interactive situation involving two or more individuals. The individuals are called players. The behavior of each player is described by a “utility function,” which gives a quantitative representation of each player’s preference for outcome. Models are then derived based upon the fundamental assumption that players make decisions which are motivated by a desire to maximize that utility function. Effective game theory representations provide clarity and insight into the factors that are at work, but may not be readily apparent when trying to examine a complex situation in detail.

Game theory can contribute to the better understanding of clinical management in the forensic hospital. The forensic hospital presents interactions and motivations which can be very distinct and more difficult to characterize than clinical practice in conventional, non-forensic settings. Additionally, the quality and nature of the players themselves are significantly more complex. Consider the common situation of a floridly psychotic man who has committed a serious crime and is now committed to the forensic hospital after being determined incompetent to stand trial. In conventional psychiatric practice, the prompt alleviation of mental symptoms would be the treatment endpoint mutually shared by the clinician and patient. In the forensic hospital setting, even though all of the measurable clinical parameters may be identical, the interaction between the clinician and the patient may be quite different. While the clinician’s goals remain unchanged, the patient may now have incentive to remain ill in order to delay confrontation of legal consequences. Furthermore, there are now additional players in the game, including the defense attorney, the prosecuting attorney, and the forensic evaluator who all shape the behavior of the clinician and patient.

A problematic but common game encountered in forensic clinical practice is malingering. The *Diagnostic and Statistical Manual of Mental Disorders, 5th Edition* (DSM-5) describes malingering as “the intentional production of false or grossly exaggerated physical or psychological symptoms, motivated by external incentives.” The DSM-5 acknowledges the propensity for malingering to occur in forensic clinical practice, and further lists criterion 1 for the diagnosis as “medicolegal context of presentation.” Indeed, data presented at the 2009 American Academy of Psychiatry and the Law annual meeting indicated that greater than 18% of patients found incompetent to stand trial were malingering their psychiatric symptoms on admission to an inpatient facility for competency restoration. This is a troubling figure given the progressively increasing demand for forensic clinical services in a system with relatively fixed resources. In California, it was estimated that in 2011, accused and convicted offenders with mental illness occupied approximately 4,500 of the 5,000 state mental hospital beds, up from 500 one decade prior.

This presentation will support developing a reductionist model of the forensic hospital malingering “game,” so that

instrumental factors at play can be clearly and directly understood. Furthermore, the model will provide a discrete and reproducible method for the clinician and hospital administrator to identify and curtail malingering, allowing scarce forensic clinical services to be appropriately and optimally utilized.

Game Theory, Malingering, Forensic Hospital

110 Extremes of Malingering: Clinicians’ Stories of Feigned Behaviors by Inmates in Corrections Facilities

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The goal of this research is to explore the bizarre behaviors displayed by inmates believed to malingering symptoms from a clinician’s perspective.

This presentation will impact the forensic science community by highlighting key elements that clinicians can use to identify and assess clients who are incarcerated and malingering.

While serious mental illness is prevalent among individuals incarcerated in jails and prisons across the country, many clinicians believe that those who feign or exaggerate symptoms represent a large percentage of the population. The *Diagnostic and Statistical Manual of Mental Disorders, 5th Edition* (DSM-5) labels these individuals as malingers and sets the criteria for such labeling as the “intentional production of false or grossly exaggerated physical or psychological symptoms, motivated by external incentives.” The incentives for malingering in an incarcerated population may or may not be intuitive; they could include presenting a mental condition that could result in reduced culpability or lenient sentencing, obtaining better facility placement, being excused from work assignments, and receiving non-indicated medications for tranquilizing effects and/or for trade/sales.

The false presentation of symptoms can be exhibited by incarcerated individuals with and without actual mental illness, thus further complicating decisions in diagnosis, treatment, and disposition. In addition, the time spent assessing these malingers reduces the already-limited availability of services for those whose symptom presentation is genuine. While assessment tools exist for identifying individuals who malingering symptoms, they are often time-consuming to administer and may require expertise in interpreting results. As such, malingering is often identified based upon clinical judgment, and not through assessment instruments.

In the community, persons who malingering may present with symptoms that are fewer or less severe than those presented by persons who are incarcerated. Perhaps one reason for this is that patients are free to “shop” until they find a provider who will concede to their requests. However, in an incarcerated population, the ability to “shop” is highly limited. Consequently, individuals may believe that they have to exhibit as “sick” of a presentation as possible in order to persuade the clinicians that they are ill and thus achieve their secondary gain. As such, a mental health provider inexperienced in working at a jail or prison may be shocked by the extreme behaviors which some inmates exhibit in pursuit of their goals. They may be less likely to attribute such behaviors to exaggeration or feigning because of the deviance of presentation from that seen in the community.

Clinical vignettes of psychiatrists and psychologists providing treatment to persons in a correctional facility will be presented. The goal is to explore the clients' incentives for malingering and the types of behaviors displayed in pursuit of these incentives. Key elements which clinicians may use to identify and assess incarcerated clients who are malingering will be highlighted.

Malingering, Corrections, Secondary Gain

111 Current Status of Clinical Research in Correctional Settings — A Review

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After attending this presentation, attendees will understand some basic principles of clinical research in correctional settings. The encumbrances involved in the consent process and the obstacles encountered in conducting research in such settings will be discussed. This presentation will also enlist some potential advantages and gains to the subjects and to society in general.

This presentation will impact the forensic science community by pointing out ramifications and implications of conducting biomedical and psychological research on inmates. The research community's benefit will be to gain ample understanding of variations in research procedures, current trends, and thinking of conducting research and **Institutional Review Board** (IRB) governance for this population, which is considered to be vulnerable. Incarcerated individuals have very high prevalences of drug abuse, alcoholism, HIV, hepatitis, and mental illness. By being able to conduct research with this population, it is hoped to advance the field in terms of new diagnostic and treatment modalities.

Research in correctional settings has always been considered controversial. On one hand, this is a set of individuals who have lost their liberty and are therefore deemed to be unable to give informed consent; on the other hand, there is potentially a wealth of clinical information that could be discovered if researched appropriately. Inmates in correctional settings can be surprisingly agreeable and amenable to clinical research for a variety of reasons while incarcerated. Also, there are several diseases, such as Hepatitis C, HIV, and conditions such as alcoholism and substance abuse, which are rampant in these populations. These findings suggest that extensive research is needed and appropriate.

In general, clinical research requires voluntary informed consent. However, working with inmates in correctional settings raises several ethical and legal dilemmas. Since this research is working with a subset of the population who have lost significant freedoms, it has been pointed out that any consent offered with respect to clinical and experimental research cannot be completely voluntary.

While there are potentially many rewards of conducting research in correctional settings, it is critically important to have extensive and special safeguards in place in order to properly achieve this goal. For example, a specialized IRB, which understands and is sensitive to protecting the rights of incarcerated individuals, is extremely important. The IRB should be constituted by especially experienced personnel who are able to evaluate protocols and, at the same time, assure subjects' rights and safety. The formation of such a committee is vital in order to minimize coercion during the informed consent process.

Clinical Research, Correctional Settings, Patient Safety

112 The MMPI-2 Test and Response Times: Evaluating Dissembling Behavior in the Field of Forensics

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After attending this presentation, attendees will more fully comprehend and appreciate the importance and utility of the computerized version of the Minnesota Multiphasic Personality Inventory-2 (MMPI-2) test and its ability to identify dissembling behaviors in subjects who undergo forensic psychiatric testing.

This presentation will impact the forensic science community by demonstrating that dissembling behaviors have an important influence on forensic psychiatric evaluation results, and the need for reliable and valid diagnostic instruments is of the utmost importance.

In the course of their work, experts in the field of forensics must evaluate not only the validity of what has been reported to them, but also the validity of the reporting. The relationship between the subject and the forensic science specialist is influenced by a series of elements which may alter the genuineness of a diagnosis (e.g., the examiner's style, the rigid and constrictive nature of the exam, the repercussions of the outcomes of the exam, the need for the results to be reported to a third party, and the tendency of evaluated subjects to dissemble). Concealment of a syndrome represents a problem in the diagnostic process that results in an increase of false negatives. A dissembler is a person who is well-aware of the truth, but denies it by mentally managing two conflicting affirmations.

In the field of legal medicine and forensics, it is necessary to recognize the limits of subjectivity and clinical observations. The use of psychodiagnostic instruments, in these cases, appears to be indispensable. The MMPI-2 test is the principle instrument, possessing indices which are able to root out concealing behaviors in subjects who are being examined.¹

"Lying and concealing" behaviors are accompanied by verbal and non-verbal elements, including such paralinguistic clues as body language.² "The latency period" plays a particularly interesting role among the various forms of non-verbal behavior and is often considered to be a non-verbal sign that the subject is probably lying.³

Beginning with the hypothesis that one can expect a longer response time by dissemblers because of their need to "manage mental information," the goal was set to verify potential information in connection to response times on the MMPI-2 test items as an instrument that can identify concealing behaviors.

This study was conducted using the computerized version of the MMPI-2, which is able to automatically calculate response times.

Fifty-nine males and 17 females were enlisted and subdivided into four groups based on the reasons for which they underwent legal medical assessment: candidacy for a driver's license; suitability to carry a firearm; appropriateness for a particular job; and fitness to be a parent. These subjects were compared to a control group made up of voluntary subjects deemed to be clinically healthy and not influenced by ulterior advantages or interests regarding the completion of the test. This group was controlled for number, age, and level of education.

The usefulness of response times in identifying "dissemblers" from the control group was evaluated by applying the

Student's *t*-test. The variables analyzed were the clinical, control, content, and supplementary scales of the MMPI-2, in addition to the total time needed to complete the entire protocol.

Preliminary results show how the distribution of average response times is differentiated between experimental and control subjects. The experimental group, on average, needs more time, as compared with the control group on scales which analyze psychological, clinical, and behavioral constructs of the MMPI-2. These early results seem to be encouraging and indicate a need for similar studies with larger sample sizes in order to increase the statistical significance.

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MMPI 2 Test, Dissembling Behaviors, Forensic Psychiatry Evaluation

I13 Assessment of the Characteristics of Foreign Perpetrators Sent for Forensic Psychiatric Observation: A Retrospective Study

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After attending this presentation, attendees will understand the characteristics and criminal acts of foreign perpetrators sent to the Council of Forensic Medicine's Forensic Psychiatric Observation department.

This presentation will impact the forensic science community by increasing awareness about the criminal acts of foreign perpetrators by presenting the results of comparison between foreigner and Turkish perpetrators' behaviors.

Objective: The Council of Forensic Medicine of Turkey is the highest level of governmental institution that provides expert services in Turkey, with its headquarters in Istanbul. Suspects are referred for psychiatric observation and diagnosis to the Forensic Psychiatric Observation Department of the Forensic Medicine Council. There are few studies about the medicolegal problems of foreigners who came to Turkey for different reasons. Moreover, there are no studies for criminal responsibility. The purpose of this study is to characterize sociodemographic attitudes, criminal acts, and criminal responsibilities of foreigners and to identify similarities and differences between local suspects who stayed in the Forensic Psychiatric Observation Department of the Council of Forensic

Medicine, matching for age and gender.

Methods: For this study, 15,418 cases, which were sent to the Forensic Psychiatric Observation Department between 1993 and 2012, were investigated individually. Two groups were formed: the study group consisted of all foreigners who were sent to the Forensic Psychiatric Observation Department between 1993 and 2012; and the control group consisted of Turkish suspects of the same gender and similar age (± 5) as compared to the foreigners who also stayed in Forensic Psychiatric Observation Department in the same time period. In order to systematize, they were chosen from the closest pages of archival books.

Results: It was found that 60 foreign suspects (0.4%) from 26 different countries were confined between 1993 and 2012. Most frequently, the foreigners were from Iran (with 12 cases, 6 of which were accused of drug trafficking). Twenty-eight percent of both groups were female. The foreign suspects' mean age was 32.2 ± 10.6 years old at the time of the crime, whereas Turkish suspects' were 31.7 ± 10.1 years old. The foreign suspects were sent for psychiatric observation 9.7 ± 6.5 months after the crime and observed for 7.6 ± 6.1 days. On the other hand, Turkish suspects were sent for psychiatric observation 20.3 ± 29.5 months after the crime and observed for 8.6 ± 5.7 days. Marital status was similar for both groups, but the educational level of foreign suspects was higher than that of the Turkish suspects. Blue-collar workers were the largest group among foreigners; farmers were the largest group of Turkish suspects. Alcohol and multiple drug use were similar between the two groups, but diacetylmorphine (heroin) use was higher among foreigners. Sixty-eight percent of foreign suspects committed criminal acts in Istanbul, as compared to 23% of Turkish suspects. Drug trafficking (37%), homicide (30%), and extortion (13%) were the leading crimes among foreigners. Otherwise, homicide (35%), extortion (15%), and injury (13%) were the leading crimes among Turkish suspects. Thirteen percent of foreigners and 15% of Turkish suspects were reported to be without criminal responsibility due to schizophrenia and other psychotic disorders.

Conclusion: This study shows that suspects who were sent to the Forensic Psychiatric Observation Department were mostly male and young adults. Most of the crimes were committed in Istanbul. The major criminal acts were drug trafficking and homicide, and criminal responsibilities were similar between the two groups.

Turkey, Foreigners, Forensic Psychiatry

I14 Characterization of Stepfather Incest in Northern Portugal

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After attending this presentation, attendees will better understand the characterization of stepfather incest in Northern Portugal from a forensic point-of-view.

This presentation will impact the forensic science community by demonstrating the positive association between having a stepfather and the risk of sexual abuse in a household.

Stepfather incest is an increasing problem in Northern Portugal society. As the number of recombined families grows, more children become vulnerable to this type of sexual abuse. Studies have demonstrated a strong positive association between having a stepfather and the risk of sexual abuse.

The goal of this study is to contribute to the characterization of stepfather incest in Northern Portugal from a forensic point-of-

view.

A retrospective study was performed based on forensic medical reports and legal outcomes of alleged cases of stepfather incest with victims under 18 years of age (N=139) from 2004 to 2011.

The selected cases represent 7.2% of all suspected sexual crimes against children. Some particular characteristics responsible for exacerbating the consequences of abuse were identified in this study: a victim's young age; long duration and high reiteration of the abusive practices; and, high rates of physical and emotional violence. The degree of intrusiveness was slightly higher compared to another intra-familial study and similar to the findings of extra-familial Sexual Abuse (SA) studies. Despite this, there were no injuries at the time of Forensic Medical Examination (FME) in 64% of the cases. The absence of injuries or biological evidence may, in part, be related to the delay in reporting the case for FME (only 8.6% were performed less than 72 hours after the last alleged SA). This delay is not only due to non-disclosure or late disclosure by the victim, but it is also due to the failure to report or the late report by those to whom the disclosure is made (or who suspect the abuse). In this study, family members were the first to raise suspicion (43%) and the first to report the case (36.7%). Most cases were reported to healthcare facilities (24.5%), with healthcare professionals being the first with respect to the possibility and responsibility of detecting and correctly orientating these cases. This reporting delay, and, subsequently, the absence of injuries, explains the low number of genetic, microbiological, and toxicological studies in evidence. Most of the forensic report conclusions were unspecific, which may be due to the characteristics mentioned above. Judicial decisions were obtained in only 15.8% of cases; about half of these went to trial. By all its characteristics, stepfather incest poses a major problem to early detection and signaling, implying serious psychosocial consequences.

Despite increasing awareness that stepfather incest is a major public health problem, there continues to be relatively little information about its distribution and determinants in Portugal. This study is the first Portuguese approach to the issue, highlighting the need for further studies, in order to achieve a better understanding and the prevention of this type of abuse.

Child Sexual Abuse, Stepfather Incest, Forensic Medical Examination

I15 Forensic Objective Competence Assessment Versus Severity of Symptoms and Global Functioning Assessment for Schizoid Inpatients Based on Egyptian Law

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After attending this presentation, attendees will better understand the application of the law for mentally ill patients through objective assessment of competence to consent to treatment using forensic instruments and clinical scales.

This presentation will impact the forensic science community by demonstrating how objective assessment may evaluate and enhance the assessment of patients' decision-making capacities.

In medical ethics, the principle of a patient's self-determination obliges the physician to obtain the patient's consent prior to medical treatment.¹ Law protects individuals from medical or psychological interventions against their desires, except in certain emergency or court-ordered circumstances. The Declaration of

Madrid stresses the patient's right to be fully informed of the nature of the condition, subject to proposed diagnostic and therapeutic procedures, including possible alternatives, and the right to choose between available methods.²

One condition for valid, informed consent is the patient's competence, which may be impaired, particularly in patients with mental health problems.³ The term "competence to consent" refers to an individual's legal capacity to accept a proposed treatment, to refuse treatment, and to select among treatment options. The question of an individual's competence to consent to (or to refuse) a proposed treatment intervention requires that the individual must: (1) be provided information relevant for the decision; (2) make the choice voluntarily; and, (3) be competent to make the decision.⁴ In most cases, if the situation causes the physician to examine competence, he will proceed using his own subjective judgment and clinical experience, and he may have difficulty applying standards suggested in the literature.⁵ Physicians often evaluate competence differently.⁶

The MacArthur Treatment Competence Study (MacCAT-T) uses instruments relating to the following four legal standards: (1) to understand information relevant to the decision; (2) to manipulate the information rationally; (3) to appreciate the significance of the information disclosed about the illness and possible treatments; and, (4) to express a choice.⁷ Legal standards for competence to consent to (or to refuse) treatment do not specifically require that deficits in relevant functional abilities must be a "product" of mental illness. Schizophrenia is associated with poor insight and cognitive dysfunction, which may influence decisional capacity. However, the mere presence of schizophrenia does not in and of itself signify that the patient has impairments in legal capacities.

This study's goal is to identify the four types of cognitive abilities which are the domain of functional abilities related to legal definitions of competence to make treatment decisions.⁸

In order to comply with the relatively new legislation in Egypt to consider the presence of severity of symptoms and deterioration of functions for the involuntary admission, the Brief Psychiatric Rating Scale (BPRS) and the Global Assessment of Functioning (GAF) scale are applied.

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Assessment, Competence, Schizophrenia

116 The Association Between Opinions Concerning Competence to Stand Trial and Sanity

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This presentation will examine the correlations between opinions of competency to stand trial and legal sanity in a sample of 10,354 pretrial assessments in criminal court conducted by 346 evaluators in the Commonwealth of Virginia. The goal of this presentation is to inform about the characteristics of the defendant, the evaluator, and the crime which were associated with the outcome of these assessments, as well as the commonality and differences in terms of factors that impact conclusions of the defendants being incompetent to stand trial and/or legally insane.

The factors that were associated with these two psycho-legal opinions will be relevant to decision-making by attorneys, mental health evaluators, and those involved in forensic policy and program development. This presentation will impact the forensic science community by serving not only to inform the forensic community about specific commonalities and differences, but also about the need to better understand the impact of such overlap between distinctly different psycho-legal concepts.

Proposition: Specifically, this presentation will focus on the association between opinions of incompetency and insanity, with these two mental health issues often being evaluated in unison but researched in isolation — thereby potentially camouflaging important interplays between these two psycho-legal opinions offered to the court by forensic examiners.

Synopsis of Content: The data for the study were collected by the Institute of Law, Psychiatry, and Public Policy (ILPPP) at the University of Virginia (UVA). Virginia Code 19.2-175 requires that all mental health clinicians who conduct competency and sanity-at-the-time-of-the-offense evaluations in the Commonwealth of Virginia complete forensic training approved by the Commissioner of Mental Health. Under a contractual arrangement between UVA and the Virginia Department of Behavioral Health and Disability Services, the ILPPP offers a five-day basic forensic training program to satisfy this requirement. Successful completion of this training requires the passing of a multiple-choice examination and the submission of a work product. After evaluators meet these requirements, the ILPPP enters their information into a web-based expert directory, the Forensic Evaluation Information System (FEIS), for use by courts throughout the state. In addition to providing a directory of qualified evaluators to the courts, FEIS also serves to inform ILPPP on the needs of the courts in the state through information collected from evaluators.

Data for the current presentation were obtained through FEIS from the forensic information form, a two-page instrument that concerns: (1) the nature of the evaluation, including the number and discipline of the evaluators, the time spent on different components of the evaluation, the use of psychological tests, and the sources of information both requested and obtained; (2) the defendant's current psychiatric diagnoses, criminal charge(s), use of psychotropic medication, past psychiatric treatment, and criminal history; and, (3) the psycho-legal opinions of adjudicative

competency and sanity, both in terms of the components of competency and the prongs of the insanity defense.

Summary of Results: To date, few studies have directly analyzed the Competency to Stand Trial (CST) and Mental State at the Time of the Offense (MSO)/insanity criteria (only two, according to this research). Furthermore, these studies centered on the end opinions of the respective evaluations, and both used small samples which prevented more detailed analyses. However, in the present data set it was possible to analyze the individual criteria for CST and MSO, and it was found that all Dusky criteria and insanity criteria were highly, significantly correlated ($p < 0.001$), with most presenting moderate strength correlations ($r > 0.30$).

Conclusions: Given this large sample of pretrial evaluations of competency and sanity, it is important to identify the legal and clinical factors which impact the outcome of these assessments, and the interplay and overlap between these two mental health issues. The impacts of both are highly relevant for the most psychiatrically and intellectually impaired individuals facing trial in various state court systems, yet both reflect issues often poorly understood by those involved in both the legal and mental health communities.

Competency, Insanity, Evaluation

117 Self-Mutilative Behaviors in a Forensic Population of Children and Adolescents: Clinical Features and Relationship With Depression

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The goal of this presentation is to show how socio-demographic factors (i.e., age, number of children in the family, parental divorce, history of sexual abuse, substance abuse, suicidal attempt, and number of attempted crimes) are potential risk factors for self-mutilative behavior.

This presentation will impact the forensic science community by suggesting how adolescents who have self-mutilative behaviors have wide-ranging problems in their lives. These results also suggest that symptoms of major depressive disorder significantly heighten the risk of self-mutilative behaviors in a forensic child and adolescent population.

Introduction: Self-Mutilation (SM) is defined as deliberate self-injury to body tissue without suicidal intent. SM typically starts in adolescence. Population-based studies suggest that, even with minor SM behavior, an elaborated screening for psychiatric disorders is recommended. Despite the increasing recognition of SM, its prognostic factors and association with depression are not well-understood.

Although various risk factors have been identified for SM behavior, there are very few studies in the literature researching SM behavior in forensic children and adolescents.

Objective: The purpose of this study was to investigate the risk factors for SM behavior and to present data on a possible relationship between SM behavior and depression levels in children and adolescents who were brought for forensic examination.

Methods: This study was approved for 295 consecutive children and adolescents who were brought for judicial examination. The socio-demographic factors of alcohol and substance abuse, history of sexual abuse and suicide attempts, and self-injurious behaviors were assessed by a semi-structured questionnaire. Depression levels were assessed with the Beck Depression Inventory.

Results: The mean age of the subjects was 14.27±1.05 years (age range 10-18 years); 90.5% were between 13 and 15 years of age; 75.6% were male and 24.4% were female; 81.4% had graduated from either junior or senior high school and 21% of these had held a part-time job while attending school; 47.5% were of low socioeconomic status; and 81.4% were members of families with at least three children.

According to the statements of the subjects, 3.7% were victims of sexual abuse, 67.5% had no substance abuse, 22% smoked cigarettes, 2% used alcohol and cigarettes, and 8.5% abused at least one substance other than alcohol and cigarettes (for example: marijuana, cocaine, LSD, heroin, and toluene).

Of these subjects, 22.7% had scars from SM behavior. The prevalence of SM behavior was 20.2% for the boys and 30.6% for the girls. This study found statistically significant differences when comparing the subjects with SM against the subjects without SM in terms of: number of children in their families; whether or not their parents were divorced; whether or not they had a part-time job; a history of sexual abuse; a history of substance abuse; a history of suicide attempts; and the number of involvements in crimes. Respectively, these were: $p=0.02$, $p=0.01$, $p=0.02$, $p=0.01$, $p<0.001$, $p<0.001$, and $p<0.001$, respectively.

In terms of depression, 41.8% of subjects with SM scored ≥ 17 on the Beck Depression Inventory, whereas the score was ≥ 17 in 18.9% of the subjects without SM. There was a statistically significant difference between these two groups in terms of Beck Depression Inventory ($p<0.001$).

Conclusions: This study attempts to identify specific risk factors for SM behavior. Data was analyzed using logistic regression analysis. Statistically significant differences were found in terms of age, number of children in the family, parental divorce, history of sexual abuse, history of substance abuse, history of suicide attempt, and number of attempted crimes.

Taken together, the results of this study suggest that adolescents with SM behaviors have wide-ranging problems in their lives. These results also suggest that symptoms of major depressive disorder significantly heighten the risk of SM behaviors in a forensic child and adolescent population. The specific characteristics of these behaviors need further investigation.

Depression, Self-Mutilation, Children and Adolescents

118 Relationship Between Self-Injurious Behaviors and Levels of Aggression in Judicial Examination of Children and Adolescents

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After attending this presentation, attendees will understand that, when the aggression levels increase, self-injurious behaviors also increase in children and adolescents who have judicial problems.

This presentation will impact the forensic science community by showing how self-injury behaviors are related with aggression levels and how these behaviors may be used as a predictive validity of unstructured clinical risk assessment and associated risk factors for aggression in predicting self- and other-directed aggression.

Background: The link between aggression and psychiatric disorders has been shown in previous studies. Aggression is a behavioral pattern typically expected in children and adolescents who have judicial problems. Aggression is defined as behavior aimed at causing harm or pain, psychological harm, personal injury, or physical distraction. Aggression may be direct or indirect, active or passive, and physical or verbal. Aggression can be directed toward self as well as others. It has been reported that self-directed aggression and self-injury are interrelated. However, the current literature consists of a limited number of studies on this issue.¹⁻⁴

Purpose: The goal of this study was to examine the relationship between aggression and self-injurious behavior in children and adolescents who have judicial problems.

Methods:

- **Subjects:** This study was approved for 295 children and adolescents who were consecutively brought for judicial examination. The mean age of the subjects was 14.27±1.05 years (age range: 10-18 years); 75.6% were males and 24.4% were females; 81.4% had graduated from either junior or senior high school; and 47.5% were of low socioeconomic status.
- **Measurement:** The socio-demographic profile of subjects and data about self-injurious behavior were collected with semi-structured interviews. The aggression levels of the subjects were determined by using the Aggression Questionnaire (AQ), which is an updated form of the Buss-Durkee Hostility Inventory. AQ is a Likert scale, consisting of 34 items which measure five traits: aggressiveness/anger; hostility; verbal aggression; physical aggression; and indirect aggression. These traits are typically used both individually and also combined to create an overall aggressiveness index. A validity and reliability study of the Turkish version of AQ was developed by Can in 2002. AQ has a high degree of internal consistency as evidenced by its Cronbach's alpha value of 0.95. According to the total score of the AQ, the level of aggression has threshold definitions (≤ 29 : very low; 30-39: low; 40-44: low-medium; 45-55: medium; 56-59: high-medium; 60-69: high; and ≥ 70 : very high).
- **Analysis:** Data on the comparison of continuous variables between groups was analyzed using Students-*t* test. In this study, statistical significance was accepted as $p<0.05$.

Results: The mean total AQ score of the subjects with self-injurious behavior was 78.04±21.0. The mean total AQ score of the subjects without self-injurious behavior was 62.75±18.05 ($p<0.01$). AQ subscale scores of physical aggression, verbal aggression, aggressiveness/anger, hostility, and indirect aggression were: 16.78±6.48; 12.70±4.01; 19.03±5.05; 17.43±6.12; and 11.72±4.37, respectively, for the group with self-injurious behavior. For the group without self-injurious behavior, the scores were: 12.91±5.6; 10.54±3.52; 15.86±4.71; 14.52±10.36; and 9.61±3.14, respectively. There were significant statistical differences between the two groups with respect to their subscale scores ($p<0.01$).

Conclusions: This study indicates that, when the levels of aggression increase, self-injurious behaviors also increase in children and adolescents who have judicial problems. This finding shows that self-injurious behaviors are related with aggression

levels, and that these behaviors may be used with predictive validity in unstructured clinical risk assessment of aggression in predicting aggression directed toward self and others. Future studies are needed to investigate the relationship between self-injurious behaviors and psychopathology, particularly aggression.

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Aggression, Self-Mutilation, Children and Adolescent

119 DIA-CRA (Craving Diagnosis): Preliminary Report on a Multidimensional Craving Measuring Scale

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The objective of this preliminary study is to demonstrate a multidimensional methodology for evaluating substance craving.

This presentation will impact the forensic science community by providing an in-depth perspective of the study and knowledge of the complex phenomenon of craving, with its multiple aspects and factors, including behavioral and physical individual responses. Therefore, this tool could help to gather more useful data in order to specifically and individually address further implications on relapse prevention and therapy.

This presentation will begin with an accurate analysis of the theoretical basis of craving, including a specific literature review on this subject. This presentation will describe the creation of a multidimensional scale, aimed at evaluating the multiple aspects of craving experimentation in individuals attending a public health clinic for substance abuse.

The scale was structured with the goal of reaching an in-depth analysis of the complex factors which determine the subjective responses to craving. Therefore, this study included evaluation subscales measuring objective physical and behavioral parameters (besides oniric activity) before and after exposure to various visual stimuli evocative of craving.

Specifically, the scale includes a questionnaire about the subjective perceptions of craving, a list of subscales of objective physical and behavioral parameters, and a sequence of visual stimuli. With regard to the latter stimuli, a team of psychologists was assigned to select different pictures representing images and/or situations thought to trigger craving impulses in the examined subjects.

Since it is widely recognized that craving induces physical and behavioral modifications, a recording of these objective parameters was included, before and after the visual exposure to the described stimuli.

The subjects who were administered this scale were chosen independently from the substance as well as the type of addiction. In other words, although the specific focus was on alcohol and other drugs, multiple types of addiction were included in the administration and evaluation of this scale, and the final findings were kept separated for study and research purposes.

On the basis of these assumptions, the preliminary results will be presented and discussed, with emphasis on the fact that this was an experimental phase aimed to gain information for the purposes of validating a recently-created scale. With this perspective, each factor that emerged was considered in order to further calibrate the scale, with particular attention to the effectiveness of this experimental diagnostic tool and its subsequent implications on relapse prevention and therapy.

Craving, Evaluation, Scale

120 Sexual Abuse in Youth-Serving Organizations

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After attending this session, attendees will be familiar with estimated rates of sexual abuse in different youth-serving organizations including the family, educational institutions, youth detention, as well as religious and volunteer organizations such as the Roman Catholic Church, the Boy Scouts of America®, and the Big Brothers Big Sisters® organizations. Attendees will better understand the dynamics of sexual abuse in the context of an ongoing relationship of trust and the relevance of this behavior to understanding intervention and prevention strategies.

This presentation will impact the forensic science community by presenting a topic that holds relevance to youth organizations, investigators, forensic experts, and attorneys as it impacts practice standards, criminal prosecution, and civil litigation.

In 2012, the Oregon Supreme Court ordered the release of 855 Ineligible Volunteer (IV) files that encompassed the submissions of "P" or perversion files to the Boy Scouts of America® (BSA) National Headquarters from local councils throughout the U.S. and internationally from January 1965 through June 30, 1984. These files had been central in the civil litigation against the BSA in Portland, Oregon, which resulted in an \$18.5 million punitive judgment against the organization for sexual abuse by scout leader, Timur Dykes, in the 1980s. The content of these files as summarized in the Warren Report and the relevance of this type of volunteer registry will be discussed along with other interventions that can be used by youth-serving organizations as part of their screening, intervention, and prevention efforts.¹

Numerous high-profile cases of sexual abuse by trusted individuals in a variety of youth-serving organizations have punctuated the problem of acquaintance child abuse and the intricacies of seduction, relatedness, and longevity which uniquely characterize this particular type of sexual exploitation. The hidden quality of these events, and the organizational commitments which serve as their platform, will be examined.

Earlier stereotypes of the overtly predatory sexual offender have proven inadequate and inaccurate, often camouflaging the more subtle dynamics in which on-going relationships are used to gain sexual access to children and adolescents. Our current understanding of these dynamics and the challenges they pose for identification and intervention within a wide array of youth-serving organizations will be the focus of this presentation with an eye toward creating a research platform for further study.

Reference:

1. **Warren Report Summary** - Boy Scouts of America. <http://www.scouting.org/filestore/youthprotection/pdf/WarrenReportSummary.pdf>

Sexual Abuse of Children, Youth Serving Organizations, BSA Ineligible Volunteer Files

I21 Forensics Implications: Adolescent Sexting and Cyberbullying

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The goal of this research is to alert forensic psychiatrists to the extensive prevalence of sexting and cyberbullying and what various roles they may play in the legal context.

This presentation will impact the forensic science community by educating attendees about the prevalence of adolescent sexting and cyberbullying and the potential legal consequences.

Adolescence is marked by establishing a sense of identity, core values, a sense of one's relationship to the outside world, and heightened peer relationships. In addition, there is also risk-taking, impulsivity, self-exploration, and a dramatic increase in sexuality. The dramatic increase in the use of cell phones and the internet has additional social implications of sexting and cyberbullying. Sexting refers to the practice of sending sexually explicit material, including language or images, to another person's cell phone. Cyberbullying refers to the use of this technology to socially exclude, threaten, insult, or shame another person. Studies of cell phone use in the 21st-century report well over 50% of adolescents use them and that text messaging is the communication mode of choice. Studies also show a significant percentage of adolescents send and receive sex messaging, both text and images. This presentation will review this expanding literature. Various motivations for sexting will also be reviewed.

Bridging the gap between adolescents and adults has forever been a societal challenge. Characterized by a period of individuation, rebellion, and changing bodies and hormones, adolescence can be a smooth transition or tumultuous. However, failure to understand the dangerous implications of the current trends in adolescent sexual behavior can lead to detrimental consequences for all involved. The challenge for psychiatrists is to recognize adolescent trends and the potential repercussions of these trends in order to be able to ask appropriate questions during psychiatric evaluations.

Adolescents who are at risk for victimization also are at risk for psychiatric consequences. Depression, suicide, mood disorders, adjustment reactions, and anxiety disorders are some of the potential psychiatric sequelae of falling victim to sexting and cyberbullying.

Sexting can entangle a child in the criminal justice system. The potential circulation and permanence of the image introduces a range of troubling legal questions. The legal implications are extensive and psychiatrists may play an important role in evaluation of some of these adolescents in the legal context. Currently, the charges in most states range from misdemeanors to Class B felonies. The legal implications are extensive and psychiatrists may play an important role in evaluation of some of these adolescents in the legal context. This presentation will also make suggestions on future remedies and preventative actions.

In summary, sexting is a widespread problem that requires the attention not only of parents but also of psychiatrists. More than ever, psychiatrists need to be increasingly diligent about

exploring these topics in detail with adolescent patients. Not only must psychiatrists educate patients of the potential dangers and victimization of sexting and cyberbullying, but psychiatrists must also educate parents, guardians, and the community. Preventive methods, including educational seminars and screening, would be the best approach to educating preteens and young adolescents.

Sexting, Adolescent, Forensic

I22 Anti-Androgen Treatment in Intellectually Disabled Adolescents With Sexually Deviant Behaviors: Medical and Ethical Implications

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The goal of this research is to explore the medical and ethical issues of using potentially effective anti-androgen therapy in intellectually disabled adolescent males with sexually deviant behaviors.

This presentation will impact the forensic science community by providing the mechanism for attendees to consider the use of anti-androgen treatment in this population with a greater appreciation of the medical-ethical implications.

A case presentation of an adolescent male, who has mild Intellectual Disability (ID) with sexually deviant behaviors, who was treated with leuprolide will be the premise of this presentation. An exploration of the medical risks and ethics of employing anti-androgen therapy with this population will be discussed. Sexually deviant behaviors have been reported in individuals with ID; such persons are more likely to commit acts against prepubescent male children when compared to perpetrators without ID. Juveniles in general comprise approximately one-quarter of all sexual assault offenders as published by the National Incident-Based Reporting System. Juvenile sex offenders, specifically those who rape and molest victims, are viewed as serious juvenile delinquents and thus subject to punishment as well as clinical treatment (e.g., multisystemic therapy, cognitive behavioral therapy, family therapy, and occasionally medications such as selective serotonin reuptake inhibitors). Anti-androgen treatment has been successful in reducing aberrant sexual behaviors among some adult sex offenders, including those with ID. Adolescents with ID who engage in sexually deviant behaviors may be unable to control their overwhelming impulses and have less cognitive capacity to utilize some of the available psychotherapeutic treatments. Therefore, anti-androgen treatment may be a viable treatment intervention for these individuals.

Literature regarding anti-androgen treatment has focused primarily on adult sex offenders, with very little research and opinions addressing such treatment for adolescent males with ID and sexually deviant behaviors. Numerous medical and ethical issues arise when considering anti-androgen treatment in adolescents with ID. These may include: capacity to consent or assent; parental requests for treatment of aberrant sexual behaviors that do not have the imminent potential of harming others (e.g., inappropriate hugging); and the medical implications of reducing sex hormones in adolescent males. The treatment team, patient, and family/guardian must weigh the risks with the potential benefits of reducing aberrant sexual impulses/behavior, societal protection, and lowered risk of the adolescent being punished criminally for these deviant sexual behaviors (e.g., incarceration, residential placement, and other legal ramifications).

This presentation will review the literature on sexually

deviant behaviors in intellectually disabled adolescent males and the above-mentioned medical and ethical concerns arising from anti-androgen treatment which target such behaviors.

From a humanistic perspective, such treatment may benefit this sub-population while protecting society; however, it is imperative that there be careful consideration of the medical risks and ethical issues with using anti-androgen medications in adolescent males.

Intellectually Disabled, Adolescents, Sexual Deviance

123 Combat-Related PTSD and Violence

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After attending this presentation, attendees will be able to: (1) comment on the historical context of Post-Traumatic Stress Disorder (PTSD); (2) enumerate the core features of the revised *Diagnostic and Statistical Manual of Mental Disorders*, Fifth Edition (DSM-5) criteria for PTSD; (3) highlight the psychological stages of a stress reaction; (4) briefly describe the etiological formulations of PTSD; (5) describe the relationship between combat-related PTSD and violence; and, (6) identify the legal implications of PTSD on criminal issues, such as criminal responsibility and mitigation.

This presentation will impact the forensic science community by providing an overview of the origin of PTSD, its relationship to violence, and implications for forensic assessment.

Recent epidemiological evidence has demonstrated the increased prevalence of PTSD related to combat exposure from the wars in Iraq and Afghanistan. The increase in PTSD, along with anecdotal reports of violence among combat veterans, has raised concerns regarding the relationship between PTSD and violence. In January 2008, *The New York Times* published a series of articles about veterans of Iraq and Afghanistan. The series identified and described 121 Iraq and Afghanistan veterans who committed killings or were charged with homicide after returning from active duty. Some authors argue that the military experiences in Iraq and Afghanistan have created a new breed of lethal and impulsive killers. However, the examination of data from past war campaigns (including the Vietnam war, in particular) does not support this claim. For example, data from the Bureau of Justice Survey of Incarcerated Veterans indicates that male veterans were found to be half as likely as other men to be held in prison, and a comparison of Vietnam-era veterans (1964-1973) with Post-Cold War-era veterans (1990-2004) produced nearly equal percentages of veterans serving a prison sentence for a violent offense.

Scientific evidence reveals a complex association between PTSD and violence. More often than not, other ingredients, such as features of antisocial personality disorder and substance abuse, contaminate the direct causal link between PTSD and violence. The revised DSM-5 criteria for PTSD broadens the definition of a qualifying trauma and expands and reorganizes the list of accompanying symptoms, which is expected to affect both civil litigation and criminal defense in a wide range of cases which involve trauma. In particular, the implications of the revised DSM-5 criteria for PTSD on criminal issues, such as criminal responsibility and mitigation, are likely to be affected and warrant further investigation.

Combat-Related PTSD, Post-Traumatic Stress Disorder, Violence

124 Queens (New York) Veterans Treatment Court: Client, Courtroom, and Procedural Characteristics

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After attending this presentation, attendees will be able to describe the procedural, client, and courtroom characteristics of the Queens Veterans Treatment Court as a model for therapeutic jurisprudence for veterans.

This presentation will impact the forensic science community by informing attendees on an increasingly popular diversion court model for veterans involved in the criminal justice system.

Approximately 2.2 million service personnel have served in Operation Iraqi Freedom (OIF) and Operation Enduring Freedom (OEF), with rates of PTSD among U.S. armed services personnel estimated to be as high as one in five.¹ There has been evidence that military veterans, and those with PTSD in particular, are more prone to involvement with the criminal justice system than their non-veteran peers.² Veterans are estimated to comprise 10% of the U.S. jail population and 14% of the prison population.^{3,4} Among the hypotheses invoked to explain criminal justice involvement of veterans have been increased anger and irritability due to PTSD.⁵ Moreover, in the popular perception fueled by media reports, combat experiences are linked to veterans' violent crimes.⁶ Veterans Treatment Courts (VTCs) have emerged as increasingly popular since the first such court opened in 2008.⁷ VTCs are diversion courts that seek to apply the concept of therapeutic jurisprudence to veterans who have become involved in the criminal justice system due to substance abuse and mental health issues. By accepting a guilty plea, veteran criminal defendants charged with non-violent felonies are assigned to mandated mental health and substance abuse treatment, mentorship, and vocational and social services. Due to the recent inception of VTCs, data on recidivism are not yet available. This presentation will describe the experience of the Queen's Veteran Treatment Court (QVTC), one of two VTCs in New York City. Procedures for screening and selecting clients by the district attorney and QVTC staff will be reviewed. In addition, characteristics of QVTC clients to date with regard to demographics, charges, and diagnoses will be presented and treatment modalities and social services provided to clients will be reviewed. Courtroom procedure including therapeutic jurisprudence as practiced by the judge during client courtroom appearances will be described. The presentation will place the QVTC in the context of the VTC movement nationwide. Areas of controversy regarding VTCs and the QVTC orientation to these problems will be discussed. This presentation will serve as an in-depth review of a specific example of this unique diversion court model.

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Veterans Treatment Court, Diversion Courts, Therapeutic Jurisprudence

125 Structural and Functional Brain Imaging Findings in Adult Psychopaths: A Literature Review

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After attending this presentation, attendees will have a detailed knowledge about structural and functional abnormalities in the brains of psychopaths. Attendees will also gain a general understanding of controversies in this field and critiques about reviewed studies. Findings will be presented in correlation to major areas of the brain which are affected in this condition.

This presentation will impact the forensic science community by providing neurological findings in psychopaths' brains which have been more consistently replicated in different studies and might be more reliably used in legal settings. It will also help to design future studies in areas where findings are inconsistent or not convincing, which include the caudate nucleus, hippocampus, and cingulate gyrus.

Less than one percent of people are psychopaths, yet psychopaths commit a disproportionate percentage of violent crimes (according to some studies, more than 30%). With advances of neuroimaging in recent decades, defense attorneys have begun to use neurological findings from the psychopathic brain in court. This strategy has generated controversy regarding the consistency and legal value of these findings. Some of these findings include structural abnormalities in the orbitofrontal cortex, ventromedial prefrontal cortex, amygdala, uncinate fasciculus, caudate nucleus, and hippocampus as well as functional abnormalities in the cingulate gyrus. Despite these findings, there have been very limited studies comparing these findings in psychopaths with findings from people who score low on the psychopathy check list. Furthermore, there is no consensus about the etiology of psychopathy and none of these structural or functional abnormalities have been considered to be diagnostic findings for psychopathy.

This literature review of the brain of the adult psychopath, as defined by Psychopathy Check List-Revised (PCL-R), focuses on structural, functional, and neurological findings. Excluded are studies on children and adolescents with conduct disorder. Also excluded are studies on galvanic skin response and neuropsychological findings. PubMed® was used to search for articles published in the period from 1993 to 2013. Studies were reviewed critically to identify potential reasons for different and/or inconsistent findings.

Most studies in this review used control groups comprised of people without mental illness. Only a few studies assigned patients with diagnoses of schizophrenia, borderline personality disorder, and autism to their control groups. This discrepancy

illustrates another area that has been criticized in psychopathy research, and this should be considered in future research projects.

Psychopathy, Neuroimaging, Neurological Finding

126 Effective Communication of Psychiatry and Behavioral Science to the Judicial System: How to Survive a Cross Examination

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After attending this presentation, attendees will: (1) be able to discuss general expectations when testifying in court; (2) be able to demonstrate familiarity and insight into attorneys' general strategies; (3) be able to discuss general issues in the formulation of valid responses during cross-examination; and, (4) demonstrate familiarity with general professional guidelines and issues of uniformity among experts' reports.

This presentation will impact the forensic science community by improving the quality of communication by mental health professionals who submit expert testimony to the courts.

In effectively communicating mental health information to the courts, experts in psychiatry and behavioral science must strike a unique balance. To meet the demands of the mental health and legal professions, issues including translating jargon for a potentially lay readership, avoiding over- and under-inclusion of information, and protecting test integrity without being evasive must be carefully considered. While various texts have outlined tenants of forensic report writing,¹⁻⁴ data suggest that even seasoned, board-certified evaluators do not always follow these guidelines.⁵ Moreover, even when such guidelines are strictly followed, it may be impossible to meet the needs of all disciplines.

In preparing for written testimony, the forensic mental health professional should be mindful of general guidelines for report writing within the fields of psychology and psychiatry, as well as the (potentially conflicting) perspectives of forensic readership. Moreover, in preparing for cases using mental health experts, legal professionals may benefit from awareness of these guidelines and learning to distinguish stylistic variation between reports from more substantive errors in approach or opinion.

This presentation is structured into three parts: (1) an example of a "straw man" forensic mental health report, rendering a hypothetical expert opinion on competence to stand trial, for audience review; (2) a simulated cross-examination of an expert witness on the opinion provided in the "straw-man" report; and, (3) an interactive discussion, including general recommendations on how to support one's opinions in both written and oral form, from the perspectives of both legal and mental health professionals. From a cross-disciplinary perspective, the audience will be exposed to questions and techniques commonly used on cross-examination, with the goal of promoting mindfulness when creating the written report.

In order to best accomplish the educational objectives, the processes of creating the "straw man" report and simulation of cross-examination deviate from actual practice in that the simulated attorney has significant input to the contents of the report, and the simulated expert witness has significant input to the content of the cross examination.

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Scientific Communication, Report Writing, Cross-Examination

I27 Mental Health in Migrant Schoolchildren: Teacher-Reported Behavior and Emotional Problems

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The goal of this presentation is to highlight the need for intervention regarding the mental health of non-Italian schoolchildren in the public school system for the prevention of mental illness.

This presentation will impact the forensic science community by demonstrating the importance of prevention planning for the mental health of migrant schoolchildren, which involves both parents and teachers.

The mental health of migrant children is becoming an increasingly important area of study due to the continued growth of migrant children in Italy. The Italian Ministry of Education and University Research (MIUR) and the Foundation for Multi-Ethnic Studies (ISMU) have presented a detailed background on the national school population of non-citizen Italian residents for the academic year of 2010/2011. School enrollment of migrant children increased from 59,389 in 1996/97 to 711,064 in 2010/11. Currently, migrant students make up 7.9% of the total student population in Italy. Primary schools have the highest number of migrant children.

A significant gap in promotion rates between Italian and foreign students has been observed, most notably at the secondary school level. Although this gap has closed slightly in the recent past, 30% of non-citizen students were not promoted to the next level in the academic year of 2009/2010. This is about twice the number of Italian secondary school students.

The migration process is physically and socially stressful. It may lead to mental health problems, particularly in children. There are few Italian studies which address migrant children's mental health. The goal of this study was to compare the prevalence and types of emotional and behavioral problems in migrant schoolchildren with those of Italian-born children. This study involved migrant (first- and second-generation) and native schoolchildren attending kindergarten, primary, and secondary school. A questionnaire was administered to teachers in order to gather information regarding family composition, country of birth, age upon arrival in Italy, religion, and general level of integration

outside the school setting. All teachers filled in the Teacher Self Report (TSR) for migrant and native children. The first of these items examines the child's participation in sports, hobbies, games, activities, involvement in organizations, jobs, chores, friendships, social interactions during play, independent work, and school functioning. The second section consists of 120 items related to behavioral and/or emotional problems during the preceding six months, as rated on a three-point scale. The main areas of this construct are aggression, hyperactivity, bullying, conduct problems, defiance, and violence. The following behavioral and emotional problems are also measured: aggressive behavior; anxiety/depression; attention problems; delinquent rule-breaking behavior; social problems; somatic complaints; thought problems; withdrawal; externalization; internalization; and, general problems. Lower scores on the Academic Performance and Adaptive Functioning Scales indicate lower functioning.¹ Higher scores on the Syndrome, Total Problems, and Externalizing and Internalizing Scales indicate higher levels of maladaptive behavior. The findings of the study show that teachers detect academic and adaptive problems more easily in migrant schoolchildren than in Italian-born natives, but they might possibly be less aware of their psychological structure.

The results of this study were that the migrant children reported more problems in adaptive functioning and academic performance when compared to native-born Italian children. Problems in adaptive functioning and academic performance in migrant children may be attributed to language difficulties and a lack of family involvement with the school and its educators.

The observations from this study provide a starting point for understanding the psychological structure and the more significant difficulties which migrant children may encounter. This study also underscores the importance of early detection of these problems, so that appropriate intervention strategies might possibly be formulated.

In Italy, it is necessary to develop a multicultural approach to promote the mental health of migrant children. It is also necessary to develop intervention models for appropriate and timely access to the Italian national health system (Sistema Sanitaria Nazionale Italiana (SSNI)), in consideration of the diverse cultural groups in Italy.

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Mental Health, Migrant Schoolchildren, Teacher-Reported Behavior

I28 The Treatment of PTSD in the Midst of Ongoing Trauma: Appropriate or Impossible?

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After attending this presentation, attendees will gain an understanding about current data available on the role and effectiveness of Post-Traumatic Stress Disorder (PTSD) treatment during ongoing trauma, with an emphasis on PTSD treatment in a jail setting.

This presentation will impact the forensic science community by providing information on the role of treating PTSD in

a jail setting where there can be ongoing trauma.

According to the National Comorbidity Survey, the estimated prevalence rate of PTSD in the general population in 2008 was 6.8%.¹ While many individuals with PTSD experience a single, symptom-inducing traumatic event, others face repeated and persistent trauma such as recurring physical and/or sexual violence in domestic relationships or torture during wartime. Inmates are a particularly vulnerable population and have a relatively increased prevalence of trauma and PTSD. Recent studies indicate that the rates of PTSD in the incarcerated population have ranged from 10% to 21%, although details about the traumatic events—specifically whether they pre-dated incarceration—are less clear.^{2,3} Incarceration presents a particularly stressful environment. It exposes inmates to restrictive and punitive housing, violence and victimization from other inmates and correctional officers, and deprivation of one's usual social supports. For those individuals who already suffer from PTSD, living in a correctional environment can continually expose them to traumatic events and make the provision of mental health care challenging.

There is a dearth of studies which examine the role of treating PTSD in the midst of ongoing traumas such as incarceration, combat, or even domestic violence. One available study examined the role of Cognitive Behavioral Therapy (CBT) in the treatment of traumas in incarcerated women with comorbid substance-use disorders. This study found CBT to be helpful in reducing traumatic stress symptoms.⁴ Another study examined the efficacy of exposure therapy versus communication skills training in a group of women who were victims of intimate partner violence, many of whom were still experiencing abuse. All subjects also received other forms of treatment, such as self-esteem training, cognitive restructuring, and coping techniques. This study found that all subjects benefited from treatment and demonstrated a reduction in post-traumatic and depressive symptoms.⁵

In the present study's sample of subjects, all are male patient-inmates hospitalized at the Bellevue Hospital Forensic Inpatient Psychiatric Service. Approximately 48% met criteria for the diagnosis of PTSD, with many of these individuals indicating the jail environment to be particularly stressful and traumatic. However, less than 10% of them were actually diagnosed with PTSD by their clinical teams. Findings from this study, including detailed demographic, clinical, and trauma-exposure characteristics, will be presented. These initial findings highlight the extent of traumatic exposure in this population.

Despite the limited knowledge about the role and effectiveness of PTSD treatment during ongoing trauma, the available studies indicate that recipients of treatment experience a reduction in symptoms of PTSD. This suggests that a greater emphasis on treating individuals with ongoing trauma can be valuable. Given the high rates of PTSD in inmates, interventions such as cognitive behavioral therapy, exposure therapy, and skills training are logical considerations when directing treatment for symptoms associated with ongoing trauma.

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PTSD, Trauma, Incarceration

I29 The Effects of Occupation Stress on Cognitive Performance in Police Officers

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After attending this presentation, attendees will understand the principles and mitigating factors that affect the occupational stress of police officers as well as their cognitive performance in the ability to perform their duties.

This presentation will impact the forensic science and law enforcement communities by presenting a better understanding about how occupational stress affects the cognitive performance of police officers as it relates to their working memory.

The hypothesis being investigated is whether police officers experiencing occupational stress will also suffer from adverse effects on their working memory. The mitigating factors under investigation for potentially impacting on an officer's cognitive performance are: empathy; perceived stress; resilience to stress; dissociative states; job burnout; and sleep quality. The other variables which were investigated are the officer's age, education, number of days and hours worked, court time, off-duty work, and family situations. Each of these principles and mitigating factors are perceived to have an impact on the officer's ability to think clearly, to act quickly, and to recall details within a short amount time. Higher scores on tests which measure empathy, resilience to stress, and quality of sleep correlate with better scores on memory tests. However, higher scores reported for dissociative states, burnout, and perceived stress correlate with lower performance on memory tests. This is the first survey that attempts to gauge these stress factors and impacts on cognitive memory, utilizing a survey pool of law enforcement officers at various levels of training and field experience, as well as time on the job.

Over 100 police officers in South Florida (Broward County) were asked to take part in this research study. Only 30 officers were willing to participate. Of the 30 officers, only 17 actually completed the surveys and working memory tests. The police officers were divided into three groups: junior officers ranging from 0 to 7 years of service; veteran officers ranging from 7 to 14 years of service; and senior officers with 14 or more years of service. The officers' responses to the survey questions and working memory tests were anonymous in order to allow for greater disclosure, more accurate answers on the surveys, and possibly improved performance of the memory tests. The officers who participated in this study completed the surveys and working memory tests on two different occasions. The first occasion was on the first day of the officers' two-week work schedule, in order to establish baseline behaviors. The second occasion was on the last day of the officers' two-week

work schedule.

The results of the surveys and memory tests indicate that the junior and veteran officers showed signs of memory failure on baseline assessment with points ranging from 4 to 17 out of 36 possible points on the memory test. The senior officers showed a considerable increase in baseline memory test scores, ranging from 15 to 25 out of 36 points. On the testing at the end of the officers' second work week, the junior officers' scores varied with three officers scoring 32 points out of the 36 points, and two officers scoring under 17 points. The only variable with higher scores was with two officers scoring higher (17 points) on the Perceived Stress Survey. The veteran officers' scores varied as well, with points ranging from 18 to 31 out of 36 points. The two veteran officers with the lowest scores also had higher scores on the Burnout Surveys. The senior officers seemed to perform the best, with scores between 23 to 28 points on the memory tests. The only apparent factor which seems to explain these score differences is that the senior officers have more experience at recalling memory under pressure than the junior and veteran officers who perform the same duties while on road patrol.

Occupational Stress, Working Memory, Police Officer

130 "Off Label" Medical Treatments in Neuro-Cognitive Disorders: Competency of the Patient and Validity of Informed Consent

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After attending this presentation, attendees will recognize characteristics in the use of "off label" medical treatments in the setting of clinical neuro-cognitive disorders.

This presentation will impact the forensic science community by discussing the competence of patients with neuro-cognitive disorders to consent to off-label use of atypical antipsychotic drugs.

As associated with an increase in life expectancy, neuro-cognitive disorders (e.g., dementia) are increasing in prevalence.¹ National Institute of Statistical Analysis (ISTAT) data from 2009 indicate a progressive increase in the number of persons affected by dementia. This information should be considered in the context of patients who are regarded as legally competent to make their own decisions but might, in reality, lack the ability to understand medical recommendations and choices, and therefore lack the ability to give truly informed consent in the making of medical decisions.

The literature suggests that persons affected by severe and chronic degenerative diseases still possess valid levels of competence to make some or all the decisions about their treatment, so no state of incompetence should be simply presumed.^{2,3} However, the literature also emphasizes how difficult it can be for elderly patients to understand the information provided to them and the complex process of obtaining their valid consent. These difficulties are compounded, particularly in elderly persons, when the neuro-cognitive disorders (by themselves and in combination with comorbidities, particularly in the context of immediate consequences after acute events) can affect the person's level of

consciousness, cognitive state, and decisional competence.

In neuro-cognitive disorders, psychotic symptoms and behavioral dysfunctions are common during the course of illness. Behavioral changes may include apathy, agitation, anxiety, depressive symptoms, verbal aggression, irritability, sleep and appetite disturbances, hallucinations, and delusions.⁴ Atypical antipsychotic drugs (e.g., risperidone, olanzapine, quetiapine, and aripiprazole, in order of their medical introduction) were considered by some authorities (as of 2004) to be the preferred treatments for these behavioral disturbances when associated with dementia.⁵ However, in Italy their use in older adults with behavioral abnormalities is off-label. Medical treatments which employ drugs in such cases (off-label) are regulated by Law No. umber 94 (/August 4, 1998), which authorizes their prescription only after obtaining the patient's written, informed consent. To provide a standard model of informed consent for the off-label use of atypical antipsychotic drugs, formats have been designed for both adult persons and Parents/Guardian *ad litem*/Custodian/Administrative Support for the interdicted person, incapacitated person, or beneficiary. In these formats, detailed explanations about the nature of treatment are provided, as well as the medical condition that the treatment is intended to remedy, the usual dosage(s), the possible medical risks and benefits of taking incorrect doses, major side effects present in the data sheet, and alternative models of treatment already provided in the information sheet. Furthermore, in the forms, scientific evidences of efficacy and safety of proposed off-label treatment reported in the literature are explained. This work fills a important gap in the implementation of bioethical principles in the clinical setting.

Against this background, to assess the specific grades of impairment induced by different chronic-degenerative diseases, not only of neuro-psychic type but also in terms of the cognitive and affective capacities involved in the decision-making process, this research has widened the clinical field to conducting such research.

This presentation describes the preliminary results of research conducted in different operative units (centers for the care of patients with Alzheimer's and other neuro-cognitive disorders and residential rehabilitation facilities for patients with psychotic disorders) involving 300 patients per unit over a three-year period.

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Neuro-Cognitive Disorders, Antipsychotic Drugs, Informed Consent

131 Assessment of Capacities to Make Treatment Decisions in Patients Affected by Neuro-Psycho-Organic Diseases

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After attending this presentation, attendees will recognize capacities to make treatment decisions in patients affected by neuro-psycho-organic diseases.

This presentation will impact the forensic science community by demonstrating how competence of patients is affected by chronic-degenerative diseases of neuro-psychic and organic natures. The goal is to also assist attendees to understand patients' medical conditions, needs for treatment, and the effects of proposed drugs and/or non-pharmacological treatments.

Obtaining informed consent is essential before performing any diagnostic-therapeutic procedure and, in general, is a fundamental element of the doctor-patient relationship. It is clear that, for the patient to be competent to give valid consent, she/she must first be properly informed about his/her health condition(s), understand the risks and benefits of the therapy, be aware of alternative treatments, and be capable of making a decision.

In regard to this last capacity, although there are different definitions in the medical and legal fields, recent scientific literature has focused on four functional aspects which underlie the assessment of a patient's *competence* to give informed consent to treatment: (1) the patient's capacity to *understand* the relevant elements of the medical condition and all the information serving to make a therapeutic choice; (2) the patient's capacity to use and *appreciate* this information, and to apply it to her/his current clinical condition (e.g., the likely consequences); (3) the capacity to *reason* adequately, organizing the available information in a logical, *rational* process (assessing pros and cons) so as to assess the possible therapeutic alternatives; and, (4) the capacity to *express* a choice.^{1,2} For the patient's consent to be valid, all of the above clinical capacities must be present.

Tools have been developed to help assess the levels of competence to give consent. Among these, the best known is the semi-structured interview, the MacArthur Competence Assessment Tool for Treatment (MacCAT-T) or the matched version for research protocols MacArthur Competence Assessment Tool for Clinical Research (MacCAT-CR).² The problem of correctly informing and obtaining valid informed consent from patients with neurocognitive diseases and patients with severe terminal-stage diseases is a hot topic at the present time. To assess the specific grade of impairment induced by different chronic diseases on the cognitive and affective capacities involved in the decision-making process, this study decided to widen the clinical field to include the specific impairments induced by: (1) chronic diseases of neuro-psychic origin; (2) chronic-degenerative diseases (including patients with systemic diseases in terminal, end-stages, patients with Alzheimer's Disease, and patients with chronic psychotic diseases); and, (3) variable clinical settings (hospice for terminal diseases, care centers for patients with Alzheimer's Disease, and residential rehabilitation facilities for patients with chronic psychotic disorders). In short, these clinical ambits are different, and each features exclusive, unique characteristics which may be appreciated by specific specialists. However, they will be coordinated and united by equally specific specialists and by the

equally specific skills of medicolegal specialists so as to attain the same goal. This presentation describes preliminary results.

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End of Life Care, MacCAT-T, Consent

132 Ethical Reflections of Compulsory Health Treatment

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After attending this presentation, attendees will be able to recognize characteristics and the nature of consent for patients with respect to Compulsory Health Treatment.

This presentation will impact the forensic science community by increasing attention given to the protection of persons with psychiatric disorders subject to protection.

The recent Conference of Regions of Italy, 30 years after Law No. 833/78 came into force, has led to reflection on the subject of psychiatric emergency treatments occurring in the absence of consensus. Law 833 of 1978 has contributed to granting mental health patients equal guarantees and equal constitutional rights which, until then, had been denied. This standpoint includes Italian Compulsory Health Treatment (TSO) for mental health patients. In accordance with Law No. 833, TSO for patients with mental illnesses categorized as serious acquired the ability to be able to treat patients under the justification of the "protection of the health of the individual." In view of the delicacy and complexities found in applying TSO and the margin for "interpretation" regarding its procedures, throughout the years a very diverse and varied understanding presents itself in terms of its application in accordance with standards, even within different branches in the same territory. And as a result of these differences, new recommendations were laid out during the recent Regions Conference.

Ordered by the constitutional laws which guarantee a person's inviolable rights, TSO finds itself positioned between individual freedom and the freedom of treatment on the one hand, and the duty to safeguard health on the other hand.

The procedure of TSO is noticeable in its various phases to provide for the many levels of safeguarding a person who is temporarily deprived of the capacity to express valid consensus. Nevertheless, the complexity of the law on TSO, as well as the interpretation margins of the procedure, have contributed to the creation of an application frame which is not free of criticism. In this context, the recommendations of the Conference of Regions have particular importance.

Here, with due respect to the recent and innovative discussions about necessary improvements to the procedures adopted in TSO, this presentation will discuss personal thoughts on the subject, specifically highlighting the increased attention given to the protection of the fundamental rights of the individual, be they minors or persons with psychiatric disorders subject to protection. Despite these new guidelines, the practical, everyday problems of providing this service remain largely ignored, regardless of if the setting is one of emergency services or, as is more frequently the

case, one where the patient is unable to consent even on a long-term basis.

Therefore, keeping in mind the psychiatrist's established responsibilities in emergency situations, this study is prepared to contribute to the subject of professional obligations at a historic moment when case law seems to assign to the psychiatrist responsibility for the "dangerousness" of the psychiatrist's patient.

This presentation will describe the preliminary results of a multicenter research study.

Law No. 833/78, Consent, Competence

133 Sexual Sadism: A Coping Strategy Model

Park E. Dietz, MD, PhD, 2906 Lafayette Road, Newport Beach, CA 92663*

After attending this presentation, attendees will have a broader and deeper understanding of the vast range of sexually sadistic behaviors, their prevalence, and their manifestations in popular culture and in the sexual behaviors of normal people and criminal offenders.

This presentation will impact the forensic science community by explaining the significance of evidence often found in home searches and in the scenes of sexual homicides.

The spectrum of sexually sadistic behaviors extends from secret fantasies, erotic tickling, and "love bites" to horrific instances of torture, mayhem, and serial murder. For over 100 years, clinicians and researchers have sought greater understanding of these behaviors without achieving much more than competing schemes of classification.¹⁻³

One of the most important observations of many early authors is that there are sexual sadists who confine themselves to imaginings, viewing natural occurrences of suffering, or writing or sketching their interests. These have been described as "ideal sadists," "symbolic sadists," or "mental sadists," and those professionals who only see sexual sadists who are seeking treatment after being found out by their wives or the police or who have committed crimes may never encounter a case and may thus conclude falsely that all sadists are criminal or dangerous.

More recent research suggests that the number of men aroused by sexually sadistic imagery who do not act criminally vastly outnumber those who do commit crimes. This is the obvious but oft missed implication of studies showing that 10% of a sample of 1,016 males had definite and/or frequent erotic response to sadomasochistic stories, that sadomasochistic themes appeared on the covers of 12% of 5,132 pornographic items for sale in a random sample of 13 "adult bookstores" in four cities, and that 9.4% of male college students had experienced whipping or beating their sexual partners, degrading their sexual partners, and torturing their sexual partners.⁴⁻⁷

Research on those participating in the Bondage/Domination/Slave/Master (BDSM) subculture have shown a similar range of sexual activities in the subcultures in West Germany, the U.S., Finland, and Canada, with no indications whatsoever of comorbid nonsexual psychopathology, and several studies suggest higher educational and occupational achievement among BDSM participants than among the general population.⁸

Studies of sexually sadistic offenders, including serial rapists and serial killers, demonstrate that these men, like other violent offenders, have psychopathology, primarily personality disorders, in addition to sexual sadism, and that many retain collections of memorabilia of their crimes.⁹⁻¹¹

Based on these and other sources, this study's thesis is that sexually sadistic fantasies and behaviors are far more common than encountered in clinical or forensic samples, and that the ways

sexually sadistic individuals cope with their offbeat sexual desires are primarily a reflection of their character and values rather than a necessary outcome of the *Diagnostic and Statistical Manual of Mental Disorders*, Fifth Edition (DSM-5) diagnosis of sexual sadism disorder. The primary coping strategies used by those who find themselves aroused by sexually sadistic imagery and activities are:

- Suppression of the thoughts and urges
- Confining the thoughts to fantasy during masturbation or sexual activity with a partner
- Use of sexually sadistic pornography for arousal during masturbation or in association with sex with a partner
- Enacting mild, symbolic, and socially acceptable sadistic activities with a consenting partner, who may or may not enjoy some or all of the activities
- Role-playing by phone or internet communication, for a fee or with an unpaid partner
- Participation in the BDSM subculture, abiding by its rules under penalty of ostracism and expulsion
- Paying a "submissive" or prostitute to submit to pre-determined sadistic acts at commercial B&D parlors, strip clubs, or brothels, which may or may not include touching or overt sexual activity
- Pressuring a sexual partner into submitting to sadistic abuse, under threat of abandonment, rejection, or replacement
- Unduly influencing a sexual partner into submitting to sadistic abuse through social isolation; psychological, physical, and sexual abuse that crushes the partner's self-esteem and sense of autonomy; and ensuring the partner remains dependent on him in as many ways as possible
- Exploiting vulnerable people (poor, young, intoxicated, addicted, developmentally disabled, emotionally unstable, or otherwise impaired) with sadistic abuse
- Committing sexually sadistic assaults through force or threat of force
- Kidnapping a victim to hold captive and abuse- and kill-at-will

It remains to be seen how often such men progress along this spectrum of coping strategies, and the extent to which life stressors or intoxication account for such progression.

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Sexual Sadism, Torture, Serial Killer

134 Why Is Mentorship Pivotal in the Future of Forensic Science?

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After attending this presentation, attendees will learn important characteristics of a good mentor. Attendees will know what characteristics to look for in potential mentors and mentees and will also develop awareness of the importance of transmission of knowledge through mentorship in their personal and professional development.

This presentation will impact the forensic science community by raising awareness on the pivotal importance of mentorship in the transmission of knowledge and overall advancement of our field.

Apropos of the 2014 American Academy of Forensic Sciences (AAFS) 66th Annual Scientific Meeting's theme of *Forensic Science Education and Mentorship: Our Path Forward*, this presentation plans to examine the multifaceted meaning of what a "mentor" is, the importance of mentorship in the personal and professional development of the Academy's (including the Section's) members and advancement of the field, as well as the personal experiences of some of the presenters, in roles both as mentors and mentees.

In Greek mythology, Mentor was the son of the divine hero, Heracles (also known as Hercules), and Asopis.¹ In Homer's epic poem, *The Odyssey*, Mentor in his old age is a friend of Odysseus. When Odysseus leaves for the Trojan War, he leaves Mentor in charge of his son, Telemachus, and of his palace. Later on, Telemachus is visited by Athena, who is disguised as Mentor.² Athena is the goddess of wisdom, courage, inspiration, civilization, law and justice, just warfare, mathematics, strength, strategy, the arts, crafts, and skill.³ Athena is also a shrewd companion of heroes and frequently influences heroic endeavors, as in the case of Telemachus. As Mentor, Athena encourages Telemachus to stand up against the suitors of his mother, Penelope, who had remained faithfully awaiting the return of Odysseus for 20 years, and to go abroad to search for his father.

The personal name, "Mentor," has been adopted in English as a term to mean someone who imparts wisdom to and shares knowledge with a less experienced colleague. This connotation of the term is due to Mentor's relationship with Telemachus and to Athena's encouragements and practical advice (while disguised as Mentor) to help Telemachus deal with his personal dilemmas.⁴

In recorded modern literature, the first recorded usage of the term "mentor" can be traced to a 1699 book entitled *Les Aventures de Télémaque* by the French writer, François Fénelon. In the book, the lead character is Mentor, and the modern application of the term is used.⁵

In the modern academic world, the term "mentorship" refers to the developmental relationship between a more

experienced mentor and a less experienced partner, who is termed the mentee or protégé. Good mentors are simply people with the qualities of good role models; they listen, guide, educate, provide insight, are practical, are accessible, criticize constructively, are supportive, and are specific. They care, they have success, and they are admirable.⁶

This presentation will include a panel of mentors and mentees. The mentors are prominent AAFS members who will discuss their personal experiences, including what to look for and how to choose a mentee, as well as their satisfactions and frustrations with the mentor role. The mentees will discuss how they chose a mentor, as well as their personal experiences, satisfactions, and frustrations with the mentee role.

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Mentor, Mentee, Knowledge

135 Assessing Group Therapy for Parricides

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After attending this presentation, attendees will either gain or reinforce a conviction of the power of group treatment for patients facing severe challenges and will be able to convey this experience in their own teaching.

This presentation will impact the forensic science community's teaching members by showing the positive influence on future generations through their attentive supervision. It will impact the broader forensic science community by illustrating through example the value of group therapy to the quality of care for forensic patients.

Background: Parricide, the killing of a parent, is rare among crimes, accounting for only 2% of homicides. Thus, it was surprising to find some 15 patients with this history in a maximum-security forensic hospital in Connecticut, which at the time was a 25-year-old, 100-bed facility. The state's Department of Mental Health and Addiction Services operates the hospital, which also serves some inmates (from the Department of Corrections) who pose special challenges or are nearing the end of their sentences with unresolved, serious mental health treatment needs. A literature review confirmed the impression that parricidal patients are especially challenging to treat, explaining their overrepresentation in the hospital's population. Applying convictions regarding the effectiveness of group therapy gained from early in residency training, and the continued influence of mentoring by senior community psychiatry faculty, it was decided to offer weekly group therapy specialized for willing patients with a history of parricide. The group became known as the Genesis Group.

Objective: To evaluate the subjective effectiveness of the Genesis Group, a weekly psychotherapy group for forensic inpatients who had murdered or attempted to murder a biological or psychological parent, along with demonstrating the benefits of the training received and, in turn, given by the therapists.

Method: Following **Institutional Review Board (IRB)** approval from the Department of Mental Health and Addiction Services, 12 former or current Genesis Group members participated in individual 30-minute semi-structured interviews regarding their experiences in the Group. The interview entailed 19 open-ended questions covering different aspects of the group experience. Topics included the group's size, valued therapeutic themes, participants' attitudes, group leaders' characteristics, and recommendations for improving the group's effectiveness.

Results: Analysis of completed responses began with identification of common themes elicited by each question. Following this, the answers from all questions were compared and contrasted for a rich representation of the patients' impressions and suggestions. As a whole, the responses indicated that participants felt that they had gained significantly from their experiences in the group, and they offered useful suggestions.

Conclusions: The Genesis Group appears to be effective in meeting its therapeutic goals. Participants reported feeling that they had achieved a helpful distance from, and tolerance for, the emotions related to their crime of parricide. They no longer felt themselves as defined by their past parricidal behavior. They felt at least a sense of having grasped their crime dynamics. They gained an understanding of their mental illnesses, a fitting sense of responsibility, and a fresh redefinition of themselves in terms of personality rather than criminality. Thus, the mentoring in group treatment that the therapists had received was affirmed by its application to a particularly challenging subset of forensic patients.

Parricide, Mentoring, Treatment

136 Dead Man Talking: Forensic Evaluations of People Who Are No Longer Among the Living

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After attending this presentation, attendees will have a thorough understanding of the types of cases requiring reconstruction of prior mental status based upon records. Attendees will also understand how to approach these cases, how to interpret relevant statutes and case law, how to document their findings clearly and relevantly, and how to support their psychiatric-legal opinions in depositions or court.

This presentation will impact the forensic science community by providing much-needed understanding of these types of cases and their potential legal ramifications, so that families, heirs, survivors, and victims can all be assured of a fair legal assessment of their issues.

Forensic psychiatry is the application of psychiatry to legal matters. This work usually involves a review of records and a clinical interview of a specific person in order to answer a specific psychiatric-legal question. But what happens when that psychiatric-legal question involves the thinking or behavior of an individual who is already dead?

While this premise might seem far-fetched to the uninitiated, in real life it occurs frequently. This presentation will consider various types of cases in which mental status examinations and capacities must be recreated by the forensic psychiatrist. Three real-life cases will be presented together with the clinical and legal stories, the methodology of inferring the person's mental status during life, and the application of these findings to the legal matters at hand. Statutory and case law relevant to each case will be presented.

One common scenario is that of testamentary capacity,

an evaluation (possibly posthumously) to determine if someone has (or had) the necessary mental abilities to write a will. Cases of testamentary capacity when the individual is still alive will be mentioned, but not discussed in this presentation. When persons who thought they should (and would) inherit significant amounts of money find that their presumed benefactors in actuality excluded them from the entire estate, they frequently request evaluations of testamentary capacity. Sometimes cases arise when an older, eccentric person leaves his or her wealth to a dog or to a gardener, rather than to the natural objects of his or her bounty. Sometimes people have no genetic heirs, and the survivors — neighbors, friends, and coworkers — make claims for inheritance. Many forensic psychiatrists, as well as other physicians, are likely to encounter these cases at some point in their careers, so a sound understanding of how to approach these cases is a fundamental skill for forensic psychiatrists. A case of testamentary capacity will be presented, including a discussion of the events leading up to the referral for psychiatric evaluation, the discovery materials reviewed, the statute under which the case was evaluated, the findings, and the outcome.

However, in addition to testamentary capacity, other scenarios often arise where someone's behavior or mental state has to be inferred for some legal situation. These cases require a special skill set in order to produce an appropriate and meaningful evaluation that can be effectively utilized in the specific case. One such scenario is that in which a person transfers funds or property to another person while still alive — but the person's competency at the time of the transfer later comes into question after the person is dead (or otherwise permanently incapacitated). These cases can even result in criminal charges for the inheritor, who may be thought to be a criminal or a psychopath. One such case will be presented and analyzed to demonstrate how to approach similar cases. This case utilizes principles considered in the testamentary capacity case, but with additional variables and information, including the criminal charges of the defendant.

Finally, dramatic cases may arise as a result of murder-suicide, for example, when the estate of one victim sues the estate of the other for wrongful death. One such case will be presented and discussed, utilizing the same parameters, including the history of the case, the relevant case law, and the documents and collateral information considered in assessing the case. The case will be discussed using case law from New Jersey, but similar case laws and statutes from other states will be presented. (This case is ultimately related to life insurance, so may be of particular interest to audience members working with insurance claims.)

The presentation will utilize didactic as well as group discussion formats.

Testamentary Capacity, Wrongful Death, Competency

137 Juvenile Delinquencies in Adana, Turkey

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After attending this presentation, attendees will gain an understanding about the general characteristics of juvenile delinquents, the types of crimes they commit, their familial characteristics, other factors that affect juvenile delinquency, and the relationship between crime and criminal etiology.

This presentation will impact the forensic sciences community by: (1) suggesting some reasons underlying juvenile delinquency; (2) identifying individual, environmental, and circumstantial factors associated with criminal behaviors; (3)

recommending general policies and strategies which would form a basis for the necessary preventive, protective, and remedial measures; and, (4) recommending building programs for preventing juvenile delinquency.

Juvenile delinquency poses a significant problem in Turkey, as in the rest of the world, prompting consideration of preventive actions and re-integration of juvenile delinquents back into society.¹

Juvenile offenders are at risk of chronically perpetuating cruel violence in the future. Preventive and supportive measures can be considered and implemented for juvenile delinquents in Turkey under the Child Protection Law. First, the protection of children's best interests, including measures of counseling, care, health care, and housing, delineates factors considered individually or together with views to ensure protection of the juvenile's family environment, providing means of education appropriate to the juvenile's age group and level of development, and leading them to develop their personality and sense of social responsibility.²⁻⁵

Data of children aged between 0 and 18 years brought to trial for allegedly committing crimes in Juvenile Courts 1, 2, and 3 of the Adana Courthouse between January 2004 and June 2013 and/or referred to/applying to the Provincial Directorate for National Education for measures taken by the court decision have been evaluated on a retrospective basis. Data were gathered from court decisions and from file archives of the Provincial Directorate for National Education, Directorate of Special Education and Counseling, Counseling Research Centers. Two hundred fifty-eight children, with full data on their socio-demographic characteristics, who were brought to court for alleged crimes and/or were applying for protective orders and measures to be taken by court decision, were also included in this study.

This study analyzes the relationship between the types of crimes juveniles commit, at what age and how they commit them, whether they came from migrant families, measures to be taken by court order, and the available socio-demographic data.

The majority of the children (45%) committed property crimes. There is a positive relationship between the general age at which juveniles committed crime, the types of crime, and the ways they committed crime. As the subjects aged, they shifted from property crimes toward crimes against persons' integrity and terror offenses, and they committed crimes more frequently in groups rather than individually ($p < 0.05$).

There is a significant relationship between the age and the general type of offense; in other words, the general type of offense changes significantly depending on the age. As the subjects got older, they more frequently committed crimes against persons and terror offenses ($p < 0.05$).

There is a relationship between being a migrant and the general type of crime. Among migrants, property crimes prevailed, and younger children worked on the streets and were taken by the courts under protection.

There is a significant relationship between measures taken by court decision (counseling, education, care, health care, and housing) and educational background among juvenile delinquents. As they reached higher levels of education, they were subjected to fewer measures taken by court decision ($p < 0.05$).

There is also a significant relationship between measures taken by court decision (counseling, education, care, health care, and housing) and their familial status as well as monthly income among juvenile delinquents ($p < 0.05$).

This study proposed to identify socio-demographic features which may exert direct or indirect effects on juvenile delinquency.⁶ Among those conditions which were associated with juvenile delinquency as a social problem, the economic and circumstantial parameters (e.g., living conditions) were striking.

Violence is more prevalent among children from suburban neighborhoods and who come from the lowest income

group.⁷ Circumstances under which juveniles were raised and the neighborhoods where they lived were found to be associated with the criminal act committed. The current findings suggest that the conditions of the neighborhoods and housing, and lower income levels, are risk factors for juvenile delinquency.^{2,7,8}

In conclusion, risk factors associated with family, school, community, and children should be well-defined to prevent juvenile delinquency. Interventions set into law, including the collection of data points, should be effective in order to offer practical solutions and to reintegrate juvenile delinquents back into society.

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Juvenile Delinquency, Crime Risks, Crime Etiology

I38 Crimes Against Persons' Integrity and Property Among Juveniles in Adana, Turkey

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After attending this presentation, attendees will gain an understanding about crimes committed by juveniles against property or persons, their perceptions of the crimes, articles of the relevant laws, and the impact of migration and profiles of the accused.

This presentation will impact the forensic science community in determining the underlying reasons behind juvenile delinquency, collecting the relevant records of crimes, creating reliable national and international databases, and developing national and international projects to set the ground for the appropriate preventive, supportive, and remedial measures.

A juvenile going through a period of tumult as a result of rapid biological and psychological changes during adolescence might not grasp the meaning and consequences of his or her acts.^{1,2} The Turkish Penal Code defines the types of crimes committed against property and persons along with punishments to be inflicted under its sub-sections. As per Article 31 of the new Penal Code No. 5237 of 2005, minors under the age of 12 years at the time of the criminal act do not bear criminal responsibility. Those who are above 12 but below 15 years of age at the time of

the act do not bear criminal responsibility if they do not understand the legal meanings and consequences of the acts they commit or if they lack the ability to control their behaviors/acts. If minors between the ages of 12 and 15 comprehend the acts they commit or are able to control their behaviors associated with the criminal act, the penalty to be imposed will be reduced. Those between the ages of 15 and 18 years at the time of criminal act are, again, subject to reduced penalties.³

In the case of short jail sentences for juvenile offenders between the ages of 12 and 18 years or if there are reasons requiring arrest, it is stipulated that delinquents serve their sentences or probation within the community supported by counseling, re-integration, and rehabilitative actions taken whenever necessary.^{2,4,5}

This study intends to define the types and frequency of crimes committed against property and persons in Adana and brought to trial in the Juvenile Court, their perceptions of the crimes, periods of probation, the articles of the relevant legislation, and the profiles of the accused.

Data of children between three and 18 years of age who were brought to trial for allegedly committing crimes in Juvenile Courts 1, 2, and 3 of the Adana Courthouse between January 2004 and June 2013 and/or who were referred to/applied to the Provincial Directorate for National Education for measures taken by court decision were evaluated retrospectively. Six hundred and eighty children brought to court for alleged crimes and/or applying for protection orders, and the measures to be taken by court decision, were included in this study.⁶

Three hundred and ten (45.6%) cases involved crimes against property, 20.6% were crimes against persons' body integrity, and the rest were terror crimes. Protection orders were issued for 204 children (30%) due to their ages and the fact that they worked on the streets.

There is a positive relationship between the age at which children commit a crime and the type of crime. In other words, older children committed crimes against persons and terror crimes rather than property crimes, which tended to be committed at earlier ages. There is a positive relationship between the type of crime and the perception of crime.⁹ Offenders displayed higher levels of perception of crime as their offenses shifted from those against property to those against persons and terror. There is a relationship between being a migrant and the perception of crime: migrants had lower levels of perception of crime.⁷

While 318 juvenile delinquents (46.8%) were put on trial under penal code articles such as theft and minor injury, 202 (29.7%) were tried under the articles of the Child Protection Law governing the relevant measures and penalty reductions.⁸

This study aimed to define the types of crimes committed against property and persons in Adana, perceptions of crime, periods of probation, the articles of the relevant legislation, and the profiles of the accused.

The data from this study suggest that property crimes (theft) constituted the highest rate, suggesting a possible relationship between crime and migration.⁹ Thus, migrants should be supported by rehabilitation, re-integration, and counseling services along with vocational courses. They should not be driven into terrorist networks; instead, they should be encouraged to engage in pro-social activities.

In conclusion, access to the records of crime poses a challenge in most countries. The creation of sound and reliable databases of juvenile criminal records will likely contribute to developing national and international projects.

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Juvenile Delinquency, Crime, Integral Migration

139 Juvenile Murderers and Delinquency in Adana, Turkey

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After attending this presentation, attendees will gain an understanding about the general characteristics of minors who committed murder and certain features of their association with crime, in particular, characteristics of the relationship between murder and juveniles and the role in different cultural contexts and criminal etiology.

This presentation will impact the forensic science community by establishing the relationship between murders committed by juveniles and their general characteristics so as to develop preventive responses and to prevent juvenile delinquency.

In the studies conducted on juvenile delinquency, internal migration should be addressed along with general factors leading the child to delinquency. Migration is both an affecting and affected process. It is divided into two types: "internal migration" (when it occurs from one region to another region of the same country) and "international migration" (when it occurs from one country to another).¹ There are very few studies which specifically emphasize the correlation of internal migration with juvenile delinquency in Turkey.² The phenomenon of internal migration from rural areas to cities due to unemployment, poverty, and economic reasons is also accompanied by the risk of delinquency for the young generation. If internal migration movements are regulated in Turkey with an eye toward decreasing juvenile delinquency (e.g., by developing preventive government policies to solve problems or improve conditions leading to internal migration), then downstream problems such as unemployment, poverty, and detachment of the child from the educational system may also be handled.³

Today, sharp objects are widely used in homes and workplaces for various purposes. These objects can be kept within reach, except for some special articles, and do not constitute

criminal objects except for their out-of-purpose use as set forth by law. However, these objects are used quite often for the purposes of assault, defense, and even murder.

Even though Turkish law limits ownership and possession of firearms, it is common in the surrounding region to carry or possess guns, both licensed and unlicensed. These firearms are owned for a variety reasons, such as traditional customs or vendetta, and they are easily supplied. In turn, this may lead to more frequent murder using firearms.

According to statistics issued by the Turkish Statistical Institute in 2011, based on the type of offense charged and data for persons under 18 years of age, 390 youngsters were charged with the offense of murder, 366 of whom were male and 24 female. In Adana, 18 male minors in 2011 and 17 male minors and one female minor in 2012 were charged with homicide.⁴

This study, based on data from judicial files of persons charged with the offense of murder at a young age, proposes to identify the reasons, motives, and frequency of these murders, the association with internal migration, the breakdown of these offenses in Turkey, and the demographic characteristics of defendants as well as the deceased victims.^{5,6}

Files registered under the Adana Courthouse National Judiciary Information System (UYAP) sent to the Third High Criminal Court of the Adana Courthouse between June 2007 and June 2013 and settled after June 2007 were analyzed to establish significant relationships between the reasons and motives of the murders committed by juveniles and to define the nature and characteristics of such relationships.^{7,8}

Regarding cases of murder by juveniles included in this study, 16 (70%) were committed willfully, three (15%) on a premeditated basis, one (5%) was by torturing, and one (5%) murder involved a child who could not defend her/himself physically and emotionally. In the majority of the cases, (55%) were stabbings, while 25% were firearms cases, and the rest were blunt trauma, strangulation, and fire setting.

Out of 20 youngsters, 18 (90%) were living in slums or detached houses in suburbs, and two (10%) were living in the city and flats. Families of 15 (75%) children were migrants. Of those, 13 (65%) migrated from rural parts of the southeast regions.

Analysis of the socio-demographic data of the deceased victims revealed that the average age was 24.82 ± 10.92 years (min: 12.75, max: 53.66), 19 (95%) were male, and one (5%) was female. Of the murdered victims, 10 (50%) were friends or acquaintances, five (25%) were not acquaintances, and five (25%) were parents, siblings, relatives, and girl/boyfriends.

This study explores the reasons and motives of murders committed at an early age, the frequency of these murders, the impact of migration, and the demographic characteristics of the accused and murdered, and contributes to the recognition and prevention of juvenile delinquency.

Juvenile delinquents included in this study were between 15 and 17 years of age, and 60% committed murder without premeditation based on impulsive decisions made during disputes with the victim. The major difference between childhood and adolescence is that judgment starts to mature.⁹⁻¹⁰ The high-cognition stage begins with a transition from concrete thinking to abstract reasoning, which involves conceptual learning. It is of vital importance to assess, in terms of forensic psychiatry, whether juveniles at the time of crime are capable of considering events willfully, consciously, and freely and if they are able to reason in healthy ways, even if they do not suffer from any psychiatric disorder.⁴

Migrated families may grow poorer as they are detached from productive activities, with their poverty growing deeper and deeper with time. Under harsh living conditions, children may feel responsible for maintaining their families, they may be obliged to drop out of school, they may suffer from language- and culture-

based differences, and they may feel socially marginalized. As 75% of children in this study are from migrant families, they may disproportionately experience the strain of these austere conditions.^{6,11,12}

In conclusion, there should be concentrated efforts to carry out training and rehabilitation activities to raise awareness in the community with an eye toward preventing juvenile murders and preventing the carrying of unregistered firearms and sharp objects by means of police intervention, as the use of firearms in murders is widespread.

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Juvenile Murderer, Delinquency, Internal Migration

140 The Discriminative Power of Criteria-Based Content Analysis: A Simulation Study in Children With Immediate and Delayed Interview

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The goal of this study is to assess the discriminative power of the instrument Criteria-Based Content Analysis (CBCA), the main component of the Statement Validity Analysis (SVA), used to evaluate the accounts of two groups of subjects: (1) children who have actually experienced an event, and, (2) children to whom the same event was only described. An additional objective is to verify the same power of the instrument in relation to the reports of the same two groups of children, but obtained after four months from the events experienced or described.

This presentation will impact the forensic science community by demonstrating how some of the criteria of the CBCA seem to have a significant power to discriminate only if evaluated at a later time (i.e., after four months).

Method: Two hundred interviews were analyzed using CBCA. From a total of 100 interviewed participants, 50 were subjected to a real event (simulation of a medical examination) and the remaining 50 participants were told the story about a boy/girl who had been subjected to a medical examination (this story was the same script subjected to the former 50 subjects by an actor who impersonated the physician). After four months, all the children were subjected to a second interview about what they had experienced (lived or described) previously. For the analysis, 17 of the 19 CBCA criteria were used.

Results: The following statistical analyses were used: a one-way Analysis Of Variance (ANOVA) for each item of the questionnaire; the "structure" of the discriminant function with the relative "weight" of each item in the process of division of the subjects in the two groups; a X^2 statistic for the verification of the non-dependence from the case; and an index of canonical correlation (Pearson's r) between the function itself and the dependent variable "group." The results of the study show that some of the CBCA criteria discriminated between those children who actually lived the event, as compared to those for whom the event was only described.

Conclusions: Despite the presence of some limitations of the study, the objectives were achieved, in part: a number of the CBCA criteria seemed to be able to discriminate between those who actually lived an event and those to whom the event was only described. Furthermore, some of the criteria of the CBCA seemed to have significant power in discriminating when evaluated after four months.

Minor Testimony, Child Abuse, CBCA

141 The Discriminative Power of Criteria-Based Content Analysis (CBCA): A Simulation Study in Adults

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The goal of this presentation is to demonstrate how the Criteria-Based Content Analysis (CBCA), the main component of the Statement Validity Analysis (SVA), was applied in a simulation study with the goal of evaluating whether this instrument is able to discriminate in adult testimonies between subjects who live a real event and subjects to whom the event is only described.

This presentation will impact the forensic science community by demonstrating how CBCA may be useful as some of its criteria are able to discriminate between persons who actually lived through an event and those to whom the event was only described.

Method: Ninety-five interviews were analyzed using

CBCA. Forty-five of the interviewed participants were subjected to a real event (a verbal conflict during what the participants thought was a real lecture) and the remaining 50 participants were told the story about a lecturer in heavy conflict with a person from the public (this story was the same script provided to the former 45 subjects via an actor who impersonated the lecturer). For the analysis, 17 of 19 of the CBCA criteria were used.

Results: The following statistical analyses were used: a one-way Analysis Of Variance (ANOVA) for each item of the questionnaire; the "structure" of the discriminant function with the relative "weight" of each item in the process of division of the subjects in the two groups; a X^2 statistic for the verification of the non-dependence from the case; and an index of canonical correlation (Pearson's r) between the function itself and the dependent variable "group." The results of the study show that some of the CBCA criteria discriminated between those children who actually lived the event, as compared to those for whom the event was only described.

Conclusions: Despite the presence of some limitations on the study, as well as difficulties encountered in using the criteria of the CBCA, the results suggest that the CBCA seems to have some utility, since some of its criteria were able to discriminate between those persons who actually lived an event and those for whom the event was only described.

Adult Testimony, Statement Validity, CBCA

142 Assessment of Parental Skills-Interview (APS-I) in Child Custody: A Research in Legal Separation

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The goal of this presentation is to highlight the importance of the assessment of parental skill that are based on a scientific evidence. In this regard, Camerini, Volpini, and Lopez have established an interview protocol called APS-I for the assessment of parental competence that was used in this research.¹

This presentation will impact the forensic science community by explaining the potential use of a psychological instrument called "Assessment of Parental Skills-Interview (APS-I)" which evaluates parenting ability as well as dysfunctions and suffering in family life..

Psychological and psychiatric professionals are being increasingly commissioned by the courts to assess parenting ability in cases of conflictual separation.² It is well known that within the legal and forensic system, evidence-based practices should be valid and reliable.³ However, it is important that each assessment with psycho-forensic value be characterized by scientific requirements under theoretical, methodological, and procedural profiles.⁴ In this regard, Camerini, Volpini, and Lopez have established an interview protocol called APS-I for the assessment of parental competence.⁵

The general goals of this study are, on the one hand, to contribute to the definition of strengths and limitations of the instrument and, on the other hand, to evaluate the ability of the APS-I to discriminate (in a significantly statistical way) between the levels of global parenting skills, comparing parents with consensual separation against parents with contentious separation. It was hypothesized that parents with contentious separation are less competent, according to literature on conflicts between ex-spouses involving an impasse in the redefinition of functions and parental responsibilities.⁶ These conflicts can involve their children and are an important risk factor to the minor, with the possibility of impacting

children's mental health and behaviors.^{7,8}

Fifty-three interviews with separated parents were administered (27 with consensual separation and 26 with contentious separation, with the duration of separation ranging from three months to 15 years); all were parents of minor children (68.7% mothers and 31.3% fathers).

The APS-I is an instrument assessing specific behaviors which define the basic functions related to a concrete parental exercise exploring three main areas: (1) social support and organizational capacity (Scale A); (2) protection (Scale B); and, (3) and warmth and empathy (Scale C). Use of the APS-I presents 24 items to the parent in a conversational manner; the scores for each item are given, through the double-blind method on a Likert scale from 1 (optimum capacity) to 5 (capacity is not exercised). The answers are evaluated according to two criteria: the ability to understand; and narrative and reflective capacity.⁹

The study resulted in the following findings: (1) the APS-I is able to discriminate the levels of parental competence between the two groups, meeting the criteria of specificity and sensitivity; (2) parents with contentious separation were less competent, both in terms of global parental ability ($F(5,1)=4,16$; $p<0.05$) and in the specific area of "protection" ($F(5,1)=7,21$; $p<0.05$); and, (3) socio-demographic variables, including education, income, and being a mother or a father, do not significantly influence parenting capacity. Only in the group of parents with consensual separation did: (1) the variable of "presence of more children" appear to influence the organizational capacity; and, (2) the duration of the separation appear to decrease global parenting skills and the Scale of warmth and empathy.

Consistent with scientific literature, this study demonstrates how conflicts may negatively impact parental capacity and how these parents may be unable to completely fulfill their roles as "protectors" of their children.¹⁰

When confronted with complex and dynamic family situations questioning child custody, it can be important to further study and assess tools such as the APS-I in order to conduct improved interventions in legal (forensic) and also psycho-social areas.¹¹

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Parental Ability, Legal Separation, Child Custody

I43 Forensic Psychiatric Consultation to the Child Death Review Committee of Oakland County, Michigan

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After attending this presentation, attendees will learn how a forensic psychiatrist consultant to a county child death review committee approaches his role and responsibilities on an interdisciplinary group made up of individuals representing varied backgrounds and interests (i.e., public health, education, law, medicine, criminal justice, psychology, and other behavioral sciences).

This presentation will impact the forensic science community by providing information and experiences from a forensic psychiatric perspective about the statutory origin and history of child death review committees in Michigan, the function of the Oakland County Child Death Review Committee (CDR) in particular, and examples of representative cases reviewed with emphasis on the consultative role in child suicides, accidental deaths, cases of undetermined manners of death, and homicides.

Society pays special attention to the deaths of children. Almost all cultures have an emotional investment in children and a sense of responsibility to ensure their well-being. When child deaths occur, there is a need to determine what caused the event and how it could have been prevented. At the child death review committee level, a coalition of specialists convenes to discuss all child deaths in a closed, confidential manner that is restricted from public view.

Not all child death review committees have forensic psychiatrist consultants, and not many forensic psychiatrists provide consultation to child death review committees. When present, the forensic psychiatrist serves an important function to the committee as a physician and as a specialist trained in psychiatry and forensic medicine. These qualifications allow the forensic psychiatrist to interface effectively with professionals from varied backgrounds: physicians; behavioral specialists; prosecutors; law enforcement; and others. The result of the collaborative environment serves to promote a better understanding of the circumstances of the child's death as well as an awareness of issues in fostering prevention and education in the community. Members of the CDR indicate that input from the forensic psychiatrist consultant with regard to suicide is critical for the proper assessment of such cases. Suicide is a function of a number of factors, including socioeconomic, intrapersonal and interpersonal stress, cultural mores, social forces, and physical and mental conditions.¹ Two or three brief CDR case examples will be presented to the audience to provide a basis for

discussion of the consultative role of the forensic psychiatrist.

In summary, the interdisciplinary members of the CDR represent areas of medicine, law, social services, and law enforcement, and endeavor to provide a more complete understanding of factors which contribute to child deaths. As a result of training, background, and experience, the forensic psychiatric consultant provides important and useful perspectives which may help to integrate other multidisciplinary viewpoints in the committee's work.

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Child, Death, Interdisciplinary

144 Significance of Suicide Note Incidence During Forensic Autopsies: An 11-Year Retrospective Review of Suicide Cases in Wayne and Monroe Counties, Michigan

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Based on more than a decade of autopsy data, the goal of this presentation is to convey to attendees the rate at which individuals leave their last words before committing suicide in Wayne and Monroe County, Michigan, with comparison to suicides which are absent any final communication.

In the interest of suicide determination, research, and prevention using the extensive pool of suicide note data presented here, this presentation will impact the forensic science community by highlighting the overall knowledge and understanding of methods as well as demographics for this manner of death in one American metropolis.

There are various reasons to leave behind a suicide note, including: the expression of one's wishes; to ease the pain and suffering which one has endured; instructions to loved ones; and even the expression of anger to oneself or toward other(s). A retrospective study was conducted at the Wayne County and Monroe Medical Examiner Offices for deaths classified as suicide. A total of 2,227 cases of suicide were reported between the years 2000 and 2011. There were 536 cases (24%) accompanied by a suicide note. Suicide notes varied from a written note in 470 cases (88%), verbal threats in 50 cases (9%), text messages in six cases (1%), email or personal computer in five cases (<1%), and five cases (<1%) left a suicide note recording on audio tape/voicemail. Males had a higher incidence of leaving a suicide note in 403 cases (75%), as compared to females in 133 cases (25%). The most common age group was the middle age group of 41- to 50-year-olds, representing 122 cases (23%). There were 426 cases racially classified as White, 93 as Black, and 17 as other races. A total of 221 cases who left notes had psychiatric histories (41%), compared to 553 cases of the 1,691 cases without a suicide note (33%). Forty-three cases of the 536 with notes (8%) had a history of prior suicide attempts, as compared to 141 (8.3%) without a suicide note. The reviewed data did not reveal a significant difference in the methods used to commit suicide in light of whether or not a suicide note was left. The leading methods of suicides with notes were gunshot wounds in 279 cases (52%), followed by hanging in 154 cases (29%), drugs in 40 cases (7%),

carbon monoxide poisoning in 37 cases (7%), sharp injuries in nine cases (1.7%), multiple injuries in nine cases (1.7%), and eight (1.5%) cases were suicide by drowning. There were 840 cases of gunshot wounds from the 1,691 without suicide notes (49.7%); 468 cases involved hanging (27.7%); 86 cases of drug overdose (5%); 62 cases of multiple injuries (3.7%); 57 cases of carbon monoxide poisoning (3%); 39 cases of sharp injuries (2.3%); 34 (2%) cases of drowning; 13 (<1%) cases of thermal/smoke and soot inhalation; and other methods accounted for the remaining 6% of cases. The observed incidence of prior psychiatric history was slightly higher among individuals who left suicide notes. The incidence of prior attempts was equally distributed among people whether or not they left suicide notes. In comparison to other studies, this study did not reveal any increased incidence of violent methods among suicide cases with or without suicide notes. The incidence of suicide notes, found in the minority of suicides in this study, is consistent with published literature, though a higher rate (42%) has been noted.^{1,2} In this study, the middle-age classification (41- to 50-year-olds) was the most common age category of note-leavers, as well as the most common method (gunshot); however, it does vary from a previous publication noting 21- to 30-year-olds and hanging as the most common ages and methods, respectively, for suicides where notes were left.³

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Suicide, Note, Autopsy

145 The Eyeball Killer: Serial Killings With Postmortem Globe Eucleation

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After attending the presentation, attendees will: (1) be aware of the details surrounding an unusual series of serial killings in which the offender dissected and removed the eyes of his victims; (2) understand the various underlying psychological factors which likely played roles in the offender's actions; and, (3) understand the importance for death investigators to recognize various clues which may link one murder to another as possible serial killings.

This presentation will impact the forensic science community by highlighting the importance of forensic psychiatry and forensic pathology to death investigations in the evaluation of a unique series of serial killings.

"And if thy right eye offend thee, pluck it out."¹

The Federal Bureau of Investigation (FBI) defines "serial killings" as the "unlawful killing of two or more victims by the same offender(s) in separate events."² Many of these criminals are believed to be psychopaths, meaning they use charm, manipulation, intimidation, and violence to control others to satisfy their own selfish needs.² The motivation for their actions is complex and can involve sexual gratification, anger, or the sadistic thrill of completely

controlling another person.³ When anger is a motivation, it is often because the killer has a rage against a subgroup of people.² When the crime is sexually motivated, the killer often eroticizes violence during their development, so that violence and sexual gratification became intertwined.² The killer can show complete emotional detachment, aggression, and hostility at the same time.⁴

The body of a 34-year-old woman working as a prostitute was found lying face-up, wearing only a bra and t-shirt, which were pulled up to expose her breasts. She had a gunshot wound to the head. At autopsy, it was discovered that the globes of both eyes had been surgically removed postmortem with such precision that there was no extraneous damage to the palpebra.

Two months later, the body of a 27-year-old female prostitute was found. She was also found lying face-up, wearing only a shirt that was pulled up over her breasts. She had three gunshot wounds. Again, the eyes were cut postmortem and the globes removed.

The third victim was a 42-year-old woman working as a prostitute. She was found dead one month after the second victim, and she was completely nude. She had two gunshot wounds to the head. She had multiple stab wounds around the eyes and face, and part of an X-ACTO® knife blade was found in one of the wounds. Both globes were absent postmortem.

The police charged Charles Albright with the murders of all three victims. Various circumstantial evidence and trace evidence were utilized in his trial, at which he was found guilty and sentenced to life in prison.⁵

Intentionally-inflicted, postmortem mutilation injuries are relatively uncommon in forensic pathology practice, but their recognition can be important for a variety of reasons: that a case likely represents a homicide; that a case may represent a hate crime, a crime of passion or rage, or a serial/sociopathic killing; and/or that a series of murders may be linked to one another.⁶⁻⁸

Addressing the question of psychological motivation in these cases must include their most unique aspects: the targeting of prostitutes and the postmortem removal of the eyes. Aspects of Albright's past, which may have played roles in his homicidal behavior, including a fascination with taxidermy and eyes, as well as a disdain for prostitutes.⁵ He demonstrated features of the paraphilias of saliromania and piquerism. He also exhibited characteristics of anti-social personality disorder as well as psychopathy, marked by impulsivity, remorselessness, manipulation, a lack of empathy, callousness, and thrill-seeking behavior.⁹ The preparation and patience required to remove the eyes postmortem do not support rage killings. Instead, psychopathy, anti-social personality disorder, and various paraphilias represent a better psychological explanation for this serial killer's motivations.

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2. <http://www.fbi.gov/stats-services/publications/serial-murder/serial-murder-july-2008-pdf>
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Questioned Documents



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J1 Artificial Aging of Crystal Violet Ink Dye

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After attending this presentation, attendees will learn the effects of light on ink pigments or dyes.

This presentation will impact the forensic science community by elucidating the use of ink dating and increasing understanding of photochemical conversion and fading of ink components under daylight or other environmental conditions.

Crystal Violet (CV) is one of the most typically used dyes in ballpoint pen inks. The CV decomposition into Methyl Violet (MV) and Tetramethyl Pararosaniline (TPR) gives information about the questioned document's age. The goal of this presentation is to determine the decomposition of CV in artificial time. In order to model the natural aging process of CV, ink entries with CV have been exposed to UV light.

Artificial aging has been used to accelerate the aging process of ink in a way similar way to what occurs naturally. The amount of light and heat the questioned document is exposed to are defined carefully to ensure aging. The modeling is intended to determine the changes observed in the solvent and dye components of ink over time.

Ink entries are exposed to conditions such as light, heat, and humidity and these conditions have an accelerating effect on normal aging processes. Debates by the court related to the possibility of artificially creating older documents has lead to need to research the topic of accelerated aging for the first time in the literature. Many studies were completed in order to date questioned documents by the accelerated aging method. Although there is controversy related to artificial aging in the document examination community, methods based on artificial aging are routinely used by several laboratories in Europe and the United States.

Artificial aging of ink on documents is used for three main reasons. The first reason is to make a document look older than it really is. The second reason is to date the ink by an accelerated aging process to obtain a small portion of an aging curve. The third reason is to study the natural aging of ink by controlling environmental conditions. Modeling of natural aging by artificial aging provides a quick way of obtaining more information about the aging processes.

Ink which had only CV as a dye was produced and used in this study. Straight lines were drawn by a pen which contains this ink. Fresh ink entries and ink entries exposed to UV light (300W) during one, two, three, five, eight, and thirteen hours were analyzed.

Graphs obtained for CV, MV, and TPR exposed up to 13 hours have given crucial information about the aging of CV. The results of this study showed that CV decreased significantly at the end of the 13 hours whereas MV and TPR increased. Also, photochemical products formed and their amount increased with

time. It can be concluded that MV and TPR were the decomposition products of CV and the photochemical products were obtained with further light exposure.

Crystal Violet, Ink Aging, Daylight

J2 Determination of Physicochemical Changes in Black Ballpoint Pen by HPLC Method

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The goals of this presentation are to help attendees understand ink chemistry, ink dating, and how to best separate the ink components on a document using High-Performance Liquid Chromatography (HPLC).

This presentation will impact the forensic science community by increasing understanding of ink dating by describing the conversion and fading of ink components under environmental conditions after separation by HPLC.

Forensic document examination is an important branch of forensic science. Examination of questioned documents with suitable methods is crucial for determining the probability of document forgeries. Ink entries in the number, signature, and date on documents are the most common issues questioned by courts. Thus, ink age and structure determinations are the most frequent questions raised by the courts. Frequent questions that are asked are by whom the ink entries were written and the age of the entries.

Literature review shows that studies related with this topic in foreign countries increase day by day. Most of these studies are about the natural and artificial aging of blue ballpoint pen ink on paper. Although there are studies related to the natural aging of blue ballpoint ink, this study is a pioneer study for the artificial aging of ink entries drawn with black ballpoint pens.

Ink entries were drawn with five different black ballpoint pens and were analyzed with the help of a modified HPLC method. Fresh ink entries and ink entries exposed to light (300W) were examined during one, two, four, and six hours, respectively. A 1.2mm punching tool was used to minimize destruction while taking samples from the ink entries on the document.

As a result of six hours of light exposure, it was determined that there was formation of demethylation of the Methyl Violet (MV) family and photochemical products. It was observed that the amount of Crystal Violet (CV) decreased significantly and turned into MV and Tetramethyl Pararosaniline (TPR). It was also observed that the amount of TPR significantly increased and photochemical products formed during the artificial aging process. When the fresh ink entries were analyzed, there were no additional products observed other than CV, MV, TPR, and Victory Blue (VB). The formation of photochemical products was determined through artificial aging.

The photochemical products obtained as a result of artificial aging are evidence of whether or not a document is exposed to light. During the degradation of dyes, most of the CV turned into MV and TPR through the demethylation process. As a result of CV decomposition, the amount of MV, TPR, and other photochemical products increased significantly by means of artificial aging.

All in all, the separation of dyes in the ink formulation was completed within a shorter run time than other studies. Decomposition of dyes belonging to the MV family in black ballpoint pens was observed through artificial aging. In order to prevent document forgery, artificial aging was used to model the physicochemical processes that inks undergo while aging.

Black Ballpoint, Ink Analysis, HPLC

J3 Black Ink Analysis With Miniaturized UV-Vis Spectrometry and Capillary Electrophoresis

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After attending this presentation, attendees will understand how ink can be extracted from paper samples, studied with UltraViolet Visible (UV-Vis) spectroscopy, and separated with Capillary Electrophoresis (CE), all in one buffer solution with surfactant. Attendees will also learn how spectra and electropherograms can be analyzed using Principle Component Analysis (PCA) to find patterns and to match unknown samples with library ink profiles.

This presentation will impact the forensic science community by providing a simple, green, inexpensive, and rapid method for the effective extraction and spectral characterization of written ink. The streamlined protocol can be utilized to generate spectral and electrophoretic profiles from extracted ink for rapid ink comparison.

Conventional written ink analysis utilizes organic solvent extraction and Thin Layer Chromatography (TLC) separation which works well with organic-based ball pen ink.¹ The popularity of water-based ink such as gel pen ink and inkjet printer ink presented challenges to conventional methods due to the low solubility of the aqueous ink in organic solvent such as pyridine. The study proposes that the usage of surfactant-based buffer solutions can enhance the extraction efficiency of versatile ink samples while maintaining compatibility with downstream analytical methods.

Fifteen black ink samples from various sources, including ball-tip pens, gel pens, inkjet printer, and fountain pens, were prepared by writing or printing ink strokes on one type of white printer paper. Six holes of 1mm diameter were punched out of one stroke using the tip of a mechanical pencil before the ink was extracted in 20 μ L of 1% Sodium Laureate Sulfate (SDS) and borate buffer at a slightly basic pH, with agitation provided by a sonicator. Only 2 μ L of the extract solution were subsequently analyzed with a miniaturized UV-Vis spectrophotometer to generate a complete UV-Vis absorbance spectrum. The components of these remaining ink extracts, after dilution in the same SDS buffer, were separated using a CE method, particularly micellar electrokinetic chromatography with the same SDS borate buffer as run buffer. UV-Vis absorbances of separated components were used for signal detection with a Diode Array Detector (DAD).

After UV-Vis scanning and CE separation, the UV-Vis spectra and electropherograms were analyzed through PCA with open-source R program code. The 3D PCA plot indicated the clustering of similar type of ink samples. This novel ink analytical method utilizes the same SDS buffer for extraction, UV-Vis and CE, which facilitates the efforts for rapid profiling of various ink samples. The use of surfactant and water as solvent avoids toxic organic solvent and presents a greener alternative to conventional methods. Ink components from six hole-punches were effectively separated and spectroscopically probed with the miniaturized UV-Vis and the DAD on the CE system, which made this method a promising applicable option for practical question document examinations.

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Ink Analysis, UV-Vis, CE

J4 Do People Always Disguise Their Writing the Same? The Trilogy

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After attending this presentation, attendees will learn the most frequently used strategies for disguising one's writing, if writers tend to use the same form of disguise each time, and if the availability of deliberately disguised known specimen writing aided Forensic Document Examiners (FDE) in their examinations.

This presentation will impact the forensic science community by providing additional research data to support the hypothesis that disguised writing will contain individual characteristics that will be repeated again whenever one attempts to disguise his/her writing. This presentation will also offer data to support the benefit of collecting deliberately disguised known-specimen writing along with natural writing specimens.

Determining whether handwriting is naturally or unnaturally prepared is a common challenge for FDEs and many variables must be taken into consideration during such an examination. Some issues include the writing instrument, writing medium, nature of the signature, age of the subject(s), health of the writer, and the date of the writing. However, the final answer being sought comes down to one of three possibilities: the writing is genuine, the writing was forged (simulated, traced) by another, or the writing was deliberately disguised by the subject (often called auto-forgery or auto simulation).

Literature states that, "To a greater or lesser extent, almost all individuals have the ability to distort their handwriting or signature" so deciding whether writings or signatures were forged by another person or distorted by the subject can be difficult or even impossible.¹ Much research has been done on this topic and many of the methods commonly used to simulate writing are also used to disguise it. "Clearly, the majority of participants displayed some overlapping features between simulation and disguise, however no specific trends in features could be established to characterize each method".²

This research study focused on disguised writings and signatures prepared by subjects who intended to deny them at a later date. A deliberately disguised document was obtained from more than 50 subjects along with natural and disguised known-specimen writings. These writings were collected by three forensic document examiners from three different geographic regions of the United States. The specimens were then used in

side-by-side examinations and comparisons with their disguised writings. Strategies used by the writers to disguise their writing will be discussed. Potential sources for error in certain problematic samples will also be discussed.

The hypothesis of the FDEs conducting this study was that, like one's natural writing, a person's disguised writing will contain individual characteristics that will be repeated again and again whenever an attempt at disguise is made. Two prior research studies have supported this premise and will also be discussed. One study reported, "In almost all instances wherein a successful disguise was used, the second attempt strongly resembled the first, even though the writings were executed up to six years apart."³ Another study conducted in 2004 found "an overwhelming majority (89% of the writers) did disguise their signatures the same way each time using the same strategies." It is also hypothesized that the comparison of said characteristics, in a side-by-side examination of questioned to known writings, also like one's natural writing, can lead to the identification of a writer of disguised writing.⁴ If this theory is proven correct, it may be necessary to require that the collection of requested known exemplars include a set of "disguised" known samples as well as normal, natural writing specimens.

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2. Auto-simulation, A Study of Self-forgery versus Disguise, by Camille Foote
3. Habit Patterns in Disguised Writing, by Stephen C. McKasson and Joseph J. Lesk
4. Disguised Signatures, Random or Repetitious, by Marie E. Durina

Handwriting, Disguise, Auto Simulation

J5 The Future State of Handwriting Examinations: A Roadmap to Integrate the Latest Measurement Science and Statistics

John P. Jones II, MBA, National Institute of Standards & Technology, 100 Bureau Drive, Mail Stop 8102, Gaithersburg, MD 20899*

After attending this presentation, attendees will understand some of the key milestones that must be accomplished for the discipline to incorporate the latest advances in measurement science and statistics into the handwriting examination process.

This presentation will impact the forensic science community by summarizing the results of a two-day conference on measurement science and standards in forensic handwriting analysis where key thought leaders discussed the future of the discipline, the barriers of achieving that future, and a roadmap for the way forward.

In 2009, the National Academy of Sciences Report, *Strengthening Forensic Science in the United States – A Path Forward*, noted that "the scientific basis for handwriting comparisons needs to be strengthened."¹ The same report also noted that recent studies have increased the understanding of the individuality and consistency of handwriting and computer studies suggesting there may be a scientific basis for handwriting comparisons. Incorporating the latest quantitative assessment techniques to further enhance the discipline is not as easy as just ordering a software program from a website.

The National Institute of Standards and Technology (NIST)

hosted the Measurement Science and Standards in Forensic Handwriting Analysis Conference that took place on June 4-5, 2013, in Gaithersburg, MD. Presenters discussed the current state of forensic handwriting analysis and also shared the latest research advancements in measurement science and quantitative analysis capabilities in the discipline. Many groups collaborated with NIST to design this conference which included a facilitated session with all attendees to discuss what the future state of handwriting analysis looks like, the barriers to achieving this future state, and a general roadmap on how to get there.

The participants noted that the future state of the discipline will incorporate the use of more quantitative analysis tools during the handwriting examination process to assess and compare handwriting characteristics. Forensic handwriting examiners will employ the use of statistical models to explain the significance of their conclusions based on the uniqueness of observed and measured handwriting characteristics. However, there is considerable debate over whether these statistics should be presented to a jury and concern about how attorneys will use this information. In the future, more research involving the use of quantitative methods for examinations and statistics will be published in peer-reviewed journals which will improve the understanding of these advancements and validate examination methods. There will be a more systematic way to convert research into best practices that examiners can incorporate into their standard operating procedures. Previously established standards will be updated or validated as new technology is used to test longstanding practices.

Additional items envisioned in the future state of the discipline will be presented along with the barriers that must be addressed and next steps that should be taken to move forward. A series of recommendations to further the discipline will be presented.

Reference:

1. National Research Council. *Strengthening Forensic Science in the United States: A Path Forward*. Washington, DC: The National Academies Press, 2009

Measurement Science, Handwriting Examinations, Statistics

J6 Handwriting Examination and Human Factors: Update of the Expert Working Group on Human Factors in Handwriting Analysis

Melissa K. Taylor, BA, 100 Bureau Drive, Gaithersburg, MD 20899; and Ted M. Burkes, BS, FBI Laboratory, 2501 Investigation Parkway, Rm 2174, Quantico, VA 22135*

After attending this presentation, attendees will learn about human factors principles, the relationship between human error and human factor influences, and the benefits of documenting a process. Attendees will have an opportunity to preview the Expert Working Group's handwriting analysis process map.

This presentation will impact the forensic science community by informing forensic science practitioners, managers, and other key criminal justice system stakeholders about how error can affect handwriting analysis and gain a better understanding of the process of handwriting examination.

Human error is an inevitable part of everyday life. In most instances the results of human error are harmless and correctable, but in circumstances such as forensic analysis, where errors may lead to the loss of life or liberty, error prevention is imperative. Through the study of human factors, the forensic science community can assess the effects of human and organizational factors on the

forensic analyses and gain a deeper understanding of when and why errors occur.

The National Institute of Justice and the National Institute of Standards and Technology are sponsoring a series of expert panels to examine the effects of human factors in forensic analyses and recommend practices to reduce the likelihood of error. Each discipline-specific working group will be comprised of experts from relevant forensic disciplines, statisticians, psychologists, researchers, and other scientific experts, in addition to representatives from the legal community, professional organizations, and other identified stakeholder groups. The second working group in this series will focus on handwriting analysis.

The Expert Working Group on Human Factors in Handwriting Analysis was convened in late 2013 and will conduct a scientific assessment of the effects of human factors on forensic handwriting analysis. The working group will evaluate existing scientific or technical knowledge in forensic handwriting analysis related to errors, where information is lacking, and apply their best professional judgment to develop recommendations on how to address human factors issues. The final report will likely address all aspects of forensic handwriting analysis, ranging from the effects of the work environment to training issues and from interpretation to research into emerging technology.

The first task the working group has taken on is developing a process map that accurately depicts the analysis process and identifies critical decision points in the process. Understanding and documenting the steps in a process, their order and dependencies, and other critical pieces of information provides a common understanding of the entire process and helps to identify problem areas. Before you can improve a process, you must understand it. This presentation will include an update on working group activities and present the forensic handwriting analysis process map developed by working group members. A brief review of the findings of The Expert Working Group on Human Factors in Latent Print Analysis will be presented as well.

Handwriting Analysis, Human Factors, Process Mapping

J7 Status of Research Into Frequency Occurrence in Handwriting and Hand Printing Characteristics

Thomas W. Vastrick, BS, 522 S Hunt Club Boulevard, Ste 217, Apopka, FL 32703*

After attending this presentation, attendees will understand the importance and status of the research into frequency occurrence in handwriting and hand printing characteristics, and the opportunity to assist in its completion.

This study will have a major impact on the forensic science community by providing yet another statistical basis for the identification sciences and help insure its continued use in the United States judicial system.

In 2010, the National Institute of Justice awarded a research grant to the University of Central Florida's National Center for Forensic Science in order to determine the frequency of occurrence in certain handwriting and hand printed characteristics. The initial idea for this project began with informal discussions with several document examiners about court decisions and judicial comments stating their concern over a lack of statistical basis for the profession.

The idea was hatched to design research that would establish the frequency occurrence for handwriting and hand printing features. The design had several areas with which to deal. First, it was important to establish what kind of population sampling should be obtained. The statisticians involved in this research determined

that the proper methodology would be to use a random-based stratified population sampling. In layman's terms this means that the sampling is to approximate percentages of certain biographical data such as age range, sex, education level, handedness, location of handwriting training, and ethnicity – all based on published intrinsic and extrinsic effects on handwriting according to Huber and Headrick.¹ Second, the project needed to select a handwriting specimens form that would provide an adequate sampling for use with as small a sampling as possible in order to keep this project from becoming too large. Fortunately and with permission, this problem was solved by the use of the handwriting specimens form developed by Dr. Sargur Srihari in previous research.² Third, it was important to select the features that would be examined. The features could not be subjective in nature such as a stroke being "long" or "short" as there would be substantial disagreement between individuals as to what exactly constitutes a particular stroke as being long or short. The statisticians emphasized that the selected handwriting and hand printing features must be objective to the point that examiners would select the same answer (reliability and reproducibility of results). Several pilot studies were conducted in order to select only features that proved to be reliable and reproducible through various Attribute Agreement Analyses. Studies were conducted on use of reproductions. It was clearly shown that the use of non-original documents did not allow for adequate reliability or reproducibility. Pilot studies were then conducted using original handwriting specimens and those features in which document examiners disagreed as to the results were eliminated from the project. Examiners also conducted multiple classifications on the same specimen in order to establish internal reliability and reproducibility within one examiner. All features that are now in this project must have passed the Attribute Agreement Analyses with 100% reliability and reproducibility.

The project is now at the stage in which the collected specimens are being classified into the database. This is a very large project and assistance from the document examination community is vital for its successful completion. A review of what has been done and how the classification system works will be offered during the presentation in order to educate, and hopefully, inspire attendees.

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1. Headrick, A.M., Huber, Roy A. *Handwriting Identification: Facts and Fundamentals*, Boca Raton: CRC Press 1999.
 2. Srihari, Sargur N. PhD; Sung-Hyuk Cha, Ph.D.; Hina Arora, M.E.; and Sangjik Lee, M.S. *Individuality of Handwriting*, *J Forensic Science*, 2002 Vol 47(4) pp 856-874
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Statistics, Frequency, Handwriting

J8 Construction of a Handwriting Database of Japanese Writers

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After attending this presentation, attendees will be given an overview of the handwriting database of Japanese writers and its application to a fundamental experiment on writer identification.

This presentation will impact the forensic science community by demonstrating the validity of the database through the introduction of the findings obtained using the database.

Two-hundred and sixty-eight subjects (139 males and 129 females) participated in the data collection. Ages varied from 20 to 59 (mean age: 36.2). The subjects were requested to write 548 Kanji characters, 46 Katakana characters, 46 Hiragana characters, 52 Latin alphabet letters, and 10 Arabic numerals. Kanji characters selected for the database were characters listed in the national

list of Chinese characters in common use and characters used for Japanese prefecture and prefectural capital names. Subjects were instructed to repeat the task five times and they wrote 3,510 characters in total. The subjects were also instructed to write a character in a 1cm square box printed on a data sheet. There were 117 boxes printed on a data sheet and six sheets were assigned to one subject per task. A digital pen was used for the data acquisition and x-y coordinates of the pen tip were acquired online. Average time necessary to one task was 40 minutes. The database had x-y coordinate data and handwriting image data. Image data was acquired by scanning the data sheet at a resolution of 800 dpi. Image data can be viewed on a monitor. An operator can load and observe one writer's image data or load several writers' handwriting and compare them. The database will be of use to both the research on handwriting and the forensic document examination.

A writer identification experiment was conducted using the database. In this experiment, writer identification was accomplished using characters that were different but having the same component characters for questioned and known handwriting. All the characters used for the experiment were constructed with two components. Four construction types (divided into right and left components, divided into upper and bottom components, inside component surrounded by another component at left and bottom sides, and inside component surrounded by another component at upper and left sides) were examined. Two characters (assuming character number one and number two) per construction type were selected for the examination. The experiment was conducted using 10 writers' handwriting samples (10 subjects times 5 repetition = 50 samples per character). The x-y coordinates on the measuring points, which had been defined beforehand on the common component, were obtained and then standardized. Euclidean distance of corresponding measuring points between questioned and known samples was calculated and compared. Identification was determined as follows: Any one sample of character number one was selected as the questioned handwriting. All samples of character number two (50 samples) were defined as the known handwriting. Euclidean distance between the questioned handwriting and any known handwriting samples were calculated and compared. This procedure was conducted on all character number one samples (50 samples), where this procedure was repeated 2,500 times on one character. Then, any one sample of character number two was selected as the questioned handwriting and all character number one samples were used as the known handwriting and the same procedure as the previous case was taken. The correct identification rate was calculated and defined as the case where the smallest distance was observed between the questioned handwriting and the known handwriting of the genuine writer of the questioned handwriting. The average correct identification rate was 58%. Right and left component construction showed the highest correct identification rate. Top and bottom component construction and inside component surrounded at top and left side construction showed low correct identification rate. These results showed the comparison of the questioned and known handwriting using only a part of a character was not valid. These results also suggested that a writer paid attention to write a character in a well-balanced shape as a whole and modifying the shape of the components according to the character he or she was going to write.

Handwriting Identification, Database, Japanese Character

J9 Report Writing and Opinion Terminology

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The goal of this presentation is to assist in the ongoing discussion about how forensic document examiners should write their report, the format they should use, the language for the "examination methodology and aids" portion of the report, and the language used to report their findings.

This presentation will impact the forensic science community by helping in the ongoing discussion regarding terminology used in report writing and conclusions.

Report writing is probably one of the more difficult parts of a Forensic Document Examiner's (FDE) work. Each FDE usually develops his own report format and wording he uses in virtually every case involving handwriting and hand printing. The wording of that report and the information conveyed to the reader must be precise in every case.

The reader of the report must have a report that stands alone. The wording of the report must be sufficiently easy to understand because the FDE uses language and technical terms, and wording in the report that could otherwise be misunderstood. These technical terms must be defined in the report, and if necessary, explained by examples from the submitted writings. Any illustrations prepared to accompany the report must also be able to stand alone as illustrations in court, when and if the case goes to trial.

The purpose of the report is to explain to the recipient that the FDE received and examined certain documents (both questioned and known). When writing the report, all the submitted documents must be clearly identified and described as to whether they are questioned or known, and placed in the proper category. Each document must be described in such a way that at time of trial the FDE is able to easily locate each one and present his conclusion concerning that document.

The FDE must clearly define and describe the purpose of the requested examination(s) and what examinations are to be performed. This is accomplished by explaining the examination methodology used to examine each aspect of the writing as well as describing any technical aids used in the course of the examination.

The conclusion(s) reached by the examination(s) must be clearly reported, as well as what factors were found and considered to be of importance in arriving at a conclusion. This is accomplished by presenting, in a logical way, the conclusion(s) and the basis for that conclusion(s). Several examples of the process can be used by inserting a segment of the examined writing in his report at this place, or make reference to it as it appears in the accompanying illustration. In either case, the significance of the characteristics, quality, features of the writing, and the weight attached to it or them must be described.

Finally, the disposition of the documents after the examination is completed must be clearly described. What should be done with the exhibits? Are they being retained in the FDE file of his office or returned to the submitter? Do they need to be kept together by the submitter for possible use in making a court illustration later, or for further examination purposes?

The wording used in each section of the reports, particularly section three where it describes the "Examination Methodology and Aids," explains to the report reader how writing habits are developed and what is considered during the course of the examination and comparison to reach a conclusion based only on the evidence within the examined writing. When the examination involves alterations and interlineations, or other document issues, this portion of the report would employ different wording that describes the procedure used in each particular case.

In summary, because report writing is unique to each

FDE, the format of the report he issues in most handwriting cases and the language he typically uses is described. An example of the text concerning the examination methodology provides background information for the reader so the conclusion reached and the basis for that conclusion is better understood.

This presentation also discusses some of the elements in report writing. In any report written by a FDE, a unique report format and language has to be developed. The purpose is to present one such format and the elements of the written report.

Report Writing, Examinations, Conclusions

J10 Background, Training, and Experience of Questioned Document Examiners: Phase I — Final Report

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After attending this presentation, attendees will understand the methodology employed in the first phase of a validation study of forensic document examination that was funded by the National Institute of Justice (NIJ). Attendees will also become knowledgeable of the typical background that is characteristic of Forensic Document Examiners (FDE) who participated in this study (practicing in the United States and Canada), including educational attainment, training, certification, experience, and other aspects of their professional preparation.

This presentation will impact the forensic science community by presenting a detailed overview of the background training, experience, and qualifications of FDEs. This includes their perception of the strengths and weaknesses in training, based on their response to open-ended questions presented in the survey. This is informative for those still in training and helpful for those who offer continuing education or training courses for document examiners.

This study presents the full findings from Phase I of a national study of FDEs, sponsored by the NIJ. Selected, partial findings were presented at the American Academy of Forensic Sciences 2013 Annual Scientific Meeting while the study was still in progress. The purpose of the study is to inform and expand the extant empirical research on FDE expertise in signature analysis. In Phase I, FDEs from across the United States participated in a multi-mode (phone and web) survey to gather background information from the professional examiners and their opinions regarding the strengths and weaknesses of education and training in forensic document examination. The study survey results (e.g., background and education) included a thematic analysis of their views on training programs.

The sampling frame was derived from the contact information on file with various regional and national professional organizations in which FDE participants are members. Examiners who participated in this study were currently employed in the United States, over the age of 18, and English-speaking. Potential participants received an advance letter describing the purpose of the study at the mailing address on file with the professional organization. Two weeks after they received an advanced letter, phone interviewers at the Center for Research Design and Analysis at the University of Nevada, Reno in collaboration with researchers at Kentucky State University, contacted potential respondents to participate in the phone survey or web survey if they preferred.

The survey probed examiner background, including their educational attainment, membership in professional organizations, certifications, training, and other professional preparation. The survey also documented the type of lab in which FDEs are employed, as well as other positions that examiners have held (including specializations outside of forensic document examination). The survey also included questions regarding examiner experience in providing expert testimony and engaging in proficiency testing as part of their training. The survey concluded with open-ended questions regarding the perceived strengths and weaknesses of examiner training. Themes from these open-ended responses are summarized and presented.

The results of this survey informed Phase II, the experimental portion of the study. Phase II involves the examination of various handwriting samples in questioned documents by both FDEs and lay participants. The background information of examiners from the Phase I survey data is informative in and of itself as an indication of the contemporary background, education, and experience of FDEs who are currently practicing in the United States.

Questioned Document Examiners, Forensic Document Examiners, Qualifications

J11 Signature Type and Complexity in Questioned/Known Signature Comparison Tasks

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After attending this presentation, attendees will understand some of the principles of cognitive psychology and gain knowledge about the relationships among signature type, signature complexity, and the deployment of attention in signature comparison tasks as they relate to process and authorship decisions in a sample of handwriting comparison tasks.

This presentation will impact the forensic science community by demonstrating the importance of engaging in theoretically-based, multidisciplinary research to an understanding of the nature of the methodology and expertise in forensic document examination.

A substantial portion of forensic document examination training is devoted to signature comparisons, handwriting, and hand printing. Forensic Document Examiners (FDEs) seek those features and characteristics which may represent the document's identifying attributes. Examiners first determine the presence or absence of features, and then qualitatively assign these features some degree of evidentiary weight to reach their decisions. Examiners are trained to look for both substantial similarities and differences among writing samples, and for repeated small characteristics which may sufficiently establish that writings are clearly the work of two individuals even though they may contain many general similarities. The number and quality of these

features allow FDEs to make assertions about the authorship of the specimen and the extent of their confidence in their decisions.¹

Many current theories of attention propose that attention is based on the relationship between a bottom-up, saliency-based attentional system and a top-down, feature-specific selection mechanism. Attention is guided by relational information about the target, or information about how the irrelevant information of a non-target differs from the features of the target. Relational models of visual search demonstrate that visual attention can be guided by attending to specific feature values such as color, size, or intensity, by inhibiting attention to irrelevant features, or by directing attention to how stimuli differ. Relational models place the target in relation to its context, offering more specific (e.g., directional) information about differences.²

Tversky pointed out that most stimuli seem to be effectively described by the presence or absence of qualitative features. He and others argued that an object is represented by a set of features or attributes, and that judgments of similarity are achieved through a process of feature-matching. Tversky's "Contrast Model" systematizes this "feature" approach, and proposes that similarity depends on the proportion of features common to the two objects, and also on their unique features. Feature matching occurs by establishing differences in quality or quantity, such as differences in color or size, or the presence or absence of the features upon which the judgment is based, usually in terms of binary variables.³ This feature matching process, along with the deployment of attentional resources, is a core process of forensic document examination.

The features available for forensic evaluation are determined in part by the nature of the writing specimens. For example, compared to stylized or mixed signatures, text-based signatures may offer a greater variety of features for evaluation. Additionally, signatures vary in terms of their complexity (e.g., the number of turning points and crossing lines), their semantic content, and any number of additional features commonly recognized within the profession as indicators of the authenticity of the writing.

This paper reports findings from a national study of FDEs (supported by Award No. 2010-DN-BX-K271, National Institute of Justice, Office of Justice Programs, U.S. Department of Justice) concerning the deployment of visual attention as it relates to signature type and complexity.

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1. Lindblom, B.S. (2006). A forensic document examiner's training. In J.S. Kelly and B.S. Lindblom (Eds.) *Scientific Examination of Questioned Documents* (2ed.). (Ch. 3, pp. 15-17).
2. Becker, S. I. (2008a). Can intertrial effects of features and dimensions be explained by a single theory? *Journal of Experimental Psychology: Human Perception and Performance*, 34, 1417-1440.
3. Tversky, A. (1977). Features of similarity. *Psychological Review*, 84, 327-352.

Feature Matching, Attention, Handwriting

J12 Feature Diagnostic Value in Questioned/ Known Signature Comparison Tasks

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After attending this presentation, attendees will understand the use of eye-tracking technology to study attention- and feature-matching processes as they relate to decision-making processes in forensic document examination.

This presentation will impact the forensic science community by demonstrating the importance of engaging in theoretically-based, multidisciplinary research to an understanding of the nature of the methodology and expertise in forensic document examination.

According to Amos Tversky, most stimuli may be effectively described by the presence or absence of qualitative features. He and others argued that an object is represented by a set of features or attributes, and that judgments of similarity are achieved through a process of feature matching. Tversky's "Contrast Model" systematizes this "feature" approach, and proposes that similarity depends on the proportion of features common to the two objects as well as on their unique features. Feature matching occurs by establishing differences in quality or quantity, such as differences in color or size, or the presence or absence of the features upon which the judgment is based, usually in terms of binary variables.¹ This feature-matching process, along with the deployment of attentional resources, is a core process of forensic document examination.

Forensic Document Examiners (FDEs) are extensively trained to conduct comparisons of signatures, handwriting, and hand printing. FDEs reach their decisions by seeking those features and characteristics which may be characterized as the document's identifying attributes or characteristics. Consistent with Tversky's Contrast Model, examiners determine the presence or absence of features, whether the features are within the writer's range of variation, and then assign these features evidentiary weight. Examiners seek not only substantial similarities or differences among writing samples, but also repeated small characteristics which may be sufficient to establish clearly that writings are the work of two individuals. The number and quality of these features allow FDEs to make assertions about the authorship of the specimen and the extent of their confidence in their decisions.²

Many studies have demonstrated that FDEs are more proficient at correctly identifying or excluding signatures than are lay people.³ This suggests that compared to lay people, trained examiners should use a greater number and variety of handwriting features in reaching their conclusions about the source of questioned signatures. However, Dyer and colleagues found by using eye-tracking methodology that FDEs and lay people appeared to view signature features similarly, although the FDE opinions were more accurate than those of the lay person control group. Dyer and colleagues suggested that this finding may be due to different cognitive processes used by FDEs and lay people for evaluating questioned signatures.⁴ The findings will be discussed in the context of the examiner's extent and kind of training, education, and experience, and will be used to illustrate the ways in which cognitive psychology can contribute to an understanding of the decision-making processes of experts in the field compared to those of lay people.

This presentation describes findings from an open-ended, qualitative survey conducted as part of a national study of FDEs (supported by Award No. 2010-DN-BX-K271, National Institute of Justice, Office of Justice Programs, U.S. Department of Justice) concerning how the examiners used signature features to reach their decisions about the authenticity of signature specimens.

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1. Tversky, A. (1977). Features of similarity. *Psychological Review*, 84, 327-352.
2. Lindblom, B.S. (2006). A forensic document examiner's training. In J.S. Kelly and B.S. Lindblom (Eds.) *Scientific Examination of Questioned Documents* (2ed.). (Ch. 3, pp. 15-17).
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Feature Matching, Attention, Handwriting

J13 Semantic Content and Signature Process Identification in Single Signature Specimens

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After attending this presentation, attendees will gain knowledge about the relationship between the extent of semantic content contained in signature specimens and the use of those contextual cues in reaching conclusions about the process (e.g., natural writing vs. simulation) by which signatures are created.

This presentation will impact the forensic science community by illustrating the relationships among the semantic content of signature specimens and the use of visually available information in Forensic Document Examiner (FDE) decision making, and the importance of engaging in theoretically-based, multidisciplinary research to an understanding of the nature of the methodology and expertise in forensic document examination.

The field of forensic document examination consists of a variety of specialized tasks related to the history and preparation of questioned documents. According to Lindblom, the wide array of tasks performed by FDEs includes the ability to identify the source of handwriting and hand printing, distinguish among genuine, forged, traced, or disguised writing, to analyze inks, papers, and other substances related to documents, and other scientific or technical analyses requiring highly specialized skills.¹

These skills include the ability to sort information according to whether or not it is diagnostically relevant in identifying or eliminating an individual as the writer of a specimen. However, this determination may be impacted by cognitive factors, such as the semantic content of the signature specimen, which may influence

the deployment of the examiner's attentional resources. In his review of dozens of studies that assessed the existence and impact of confirmation bias, Nickerson highlighted two paths by which confirmation bias occurs: (1) the preferential treatment of evidence that supports existing beliefs; and, (2) the overweighting of positive confirmatory instances.² The preferential treatment of evidence that conforms to what an individual believes does not necessarily entail completely ignoring contrary information, but it has been empirically demonstrated that selective attention and selective information-seeking do occur. This suggests that the negative information is not ignored, *per se*, but is cognitively countered by means of finding information that either explains the discrepancy or invalidates it. In much the same way, the overweighting of positive confirmatory evidence may occur as a complementary process to the underweighting of disconfirmatory evidence. These findings suggest that the type of signature (e.g., text-based, mixed, or stylized) may produce different levels of bias, such that signatures which are generally more legible and thus high in the semantic content, may influence the outcome of an assessment of whether the signature in question is genuine, simulated, or disguised. Signatures that are high in semantic content may be approached in a more top-down manner, while those that are more stylized may be approached in a bottom-up fashion.

This presentation will discuss the influence of top-down vs. bottom-up processing by comparing process decisions for text-based vs. stylized single signature specimens.

Specifically, this paper discusses findings from a national study of FDEs (supported by Award No. 2010-DN-BX-K271, awarded by the National Institute of Justice, Office of Justice Programs, U.S. Department of Justice) concerning the application of cognitive theory to understanding the nature of attention, feature extraction and weighting, and decision-making in forensic document examination.

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1. Lindblom, B.S. (2006). A forensic document examiner's training. In J.S. Kelly and B.S. Lindblom (Eds.), *Scientific Examination of Questioned Documents* (2ed.). (Ch. 3, pp. 15-17).
2. Nickerson, R.S. (1998). Confirmation bias: A ubiquitous phenomenon in many guises. *Review of General Psychology*, 2, 175-220

Feature Matching, Attention, Handwriting

J14 Effective Court Charts

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After attending this presentation, forensic document examiners will learn the most effective ways to demonstrate findings in disputed document cases. Forensic document examiners learn simple and more complex technologies available to them for court chart construction.

This presentation will impact the forensic science community by improving their basic preparation of court exhibits suitable for specific types of cases.

This presentation will provide attendees with an understanding of elements present in an effective court chart. In a disputed will case, 4X enlarged questioned and known signatures could be mounted on foam board and labeled. Questioned overwriting from an altered medical record may be enlarged, printed on a standard sheet of paper, and displayed with a projector for opaque items. Images of evidence imported into popular software programs allow for the projection of significant features of questioned and known writings on a screen in the court room.

These and an array of familiar methods and new technologies that produce effective court presentations for forensic document examiners will be offered.

Forensic document examination is the application of allied sciences and analytical techniques to questions concerning documents. Disputed handwriting cases require the visual, microscopic, and instrumental analysis of questioned documents and known documents. These are then compared and an evaluation of the significance of the individualizing characteristics completes the examination. The creation of an effective demonstrative exhibit conveys significant findings to a jury. Chart preparation can also be a very important part of the examination process by scanning questioned and known writings in order to facilitate a side-by-side comparison. These charts may accompany the report and depict the significant features which support the conclusion in the case. Charts may also be utilized in the presentation of photomicrographs that are used to depict the qualities of a black ballpoint pen inked signature in order to prove that it is an original document rather than a copy.

The preparation of a simple court exhibit using a slide presentation program will be demonstrated along with standard paper exhibits meant to be published to the court. Graphic imaging software programs allow the creation of study charts that may also be attached to a report. Study charts in signature cases capture all of the questioned and known signatures, eliminating the surrounding printed material from each evidence item. A study chart arranges all of the questioned and known signatures in the case in a nutshell. Demonstrative charts can also be generated using the report features of a database that assists forensic document examiners in the collection, dissection, analysis, and presentation of forensic results.

The type of chart constructed must suit the case, the attorney, the judge, and the court facilities in order to be effective. When a courtroom is equipped with very limited technology, the most effective charts may be 4-6X enlargements of evidence mounted on foam board. Additionally, some attorneys may have their own preferences for presentation, such as the use of one enlarged questioned signature paired with one enlarged known signature on several individual paper sheets rather than the arrangement of one questioned signature with five or six known signatures on the same chart or sheet. Effective charts suitable to particular cases and situations will be demonstrated.

Charts, Court Presentation, Documents

J15 All Copies Are Problematic

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The goal of this presentation is to review the problems associated with the examination of photocopies.

This presentation will impact the forensic science community by providing information to attendees so that they will become familiar with the problems associated with the examination of photocopies.

Frequently, recipients of reports from a Forensic Document Examiner (FDE) expect the FDE to be able to determine with absolute certainty the authorship of handwriting and hand printing material or the presence of alterations or interlineations on the documents submitted for examination. Under no circumstances is this always possible. The evidence present on the document being examined limits the conclusion reached by the FDE because the conclusion must be based on the totality of the evidence present on the documents being examining.

When the document is a copy, the evidence in the writings

or other features of the copied material is always a limiting factor and the conclusion reached by the FDE is generally less than conclusive. In every case where the submitted document is a copy, the real question is whether the examined material on the copy is actually on the original the copy purports to represent. Examples of answering the real question are presented.

A number of factors determine the value of the observable evidence on copies: some are the color of the ink used to write the original material, whether a portion of the writings on the original is overwritten, or partially covered by a stamp impression, or partially obliterated by other written material, etc. These characteristics are limiting factors when examining a copy of an original on which they appear.

Trying to determine the significance of features of writing can be difficult when examining original writing. When examining the original, virtually every characteristic, quality, and feature of the writing is visible and can be examined with the aid of magnification, different wavelengths of light, changes in the angle of illumination, etc. If a copy of the original document is examined, that examination is limited to the visible elements of toner, ink jet printing, etc., on the copy that may or may not correspond to a counterpart characteristic, quality, or feature on the original.

If the examined document is a copy of a copy, or is a multiple-generation copy of the original, the accuracy of the recorded material on the copy becomes increasingly less. The new copy is the result of the copier scanning, toner, or ink jet ink applied to the document being scanned. What the scanner sees, and the accidental application of toner or ink applied to the first-generation copy or multiple-generation copy, are usually deposited on a copy and must be considered during the examination process. One example of this is random toner that may be in a location that could be misinterpreted as a critical feature on the original. For example, a dot over a staff that is supposed to be the letter "i" but the writer of the letter on the original did not place a dot over the eye staff.

A feature, such as a pencil outline or indentation of writing, is lightly impressed in the original document and may be hard to see when examining the original; these particular features, when copied, may be non-existent on the copy. Such features can be extremely significant when present in the original document. Not having these recorded characteristics on the copy being examined, the FDE may misinterpret the evidence on the copy and reach the wrong conclusion. Indented outlines on original documents are virtually never seen on the photocopy of a document.

In summary, it is not possible to reach an unqualified conclusion in every case the FDE examines. Various factors in the writings combine to limit the significance that can be attached to the characteristics, qualities, and features of the examined writing to support a conclusion that it is or is not written by the same writer. This is especially true when the examined writings are copies. The reason is that not all of the characteristics, qualities, and features of the writing on an original are present on the copy. Additionally, it can never be assumed that the writing on a copy is actually present on the original the copy purports to represent.

Copy, Interlineations, Evidence

J16 Assessment of Early Childhood Writing Development Among Family Sibling Members

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After attending this presentation, attendees will become familiar with a first-hand study into the individual development of writing characteristics assessed from three family sibling members

each separated by two years in age, grades two, four, and six. Attendees will further gain an understanding of the evaluation process conducted in the study and assessment of each of the children's writing, specifically hand printing characteristics and the stages of their development. Examples of each of the sibling's hand printing samples will be exhibited to demonstrate identifiable writing characteristics when dissected feature by feature.

This presentation will impact the forensic science community by the study of writing samples by three young siblings, educated in elementary school, assessing and understanding the development stage of the early writing habits, specifically hand printing. A review of each sibling's various writing samples and developing characteristics will also be discussed. This presentation will provide an evaluation of the sibling writing samples, the developing writing characteristics, and identifying features that discriminates between each child sibling from the other within the same family.

Handwriting is a learned behavior during elementary instruction. The learning process of writing development is a product of a young student's mental images of copybook letter forms taught in grade school. As the student matures, their writing becomes individualized and identifiable through a unique interaction of the writer's mental and physical abilities. Depending on the elementary school's educational curriculum, handwriting instruction is becoming less emphasized with regard to classroom instruction and the amount of time devoted to writing proficiency. Although there is less classroom instruction devoted to handwriting proficiency, elementary students will still be exposed to a basic handwriting structure. As the trend suggests and depending on the institution, elementary school curriculums are not providing the time to teach the cursive writing style, but rather emphasizing the hand letter or hand printing style of writing. Students exposed to learning with limited overall handwriting instruction will still develop individual hand printing habits such as basic letter construction, letter proportion, alignment, punctuation, and a personal "signature" as one matures.

As part of the procedure to a handwriting examination for comparison purposes, a forensic document examiner studies handwriting sample features to assess a writer's class and individual writing characteristics. A study will be presented on the analysis, assessment, evaluation, and noting of each sibling's writing habits as each one has progressed so far in the development of writing.

Hand Printing, Handwriting, Writing Characteristics

J17 Halftone Patterns in Security Printing

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After attending this presentation, attendees will have a basic understanding of the differences between commercial and security artwork found on printed documents, specifically the basic halftone elements used to create a continuous tone image.

This presentation will impact the forensic science community by providing better understanding of the hurdles that a counterfeiter must overcome to make a convincing document. Understanding these design techniques will help an examiner know what type of artwork is to be expected in a genuine document.

Vector-based artwork, in theory, has an infinite resolution because it uses shapes with defined curves and edges to create an illustrated image. Security printers use this type of line art for many elements of a document including guilloche patterns, patterned motifs, backgrounds, and simple objects that can be

rendered using lines and solid blocks of color. However, line art cannot create continuous tone images unless the lines themselves are used as a halftone screen element.

Halftone screening refers to various graphic arts techniques used to simulate continuous tones in printed material using a limited selection of ink colors, usually by transforming a continuous tone image into a pattern of microscopic dots that vary in their size, spacing, shape, and/or color.

Amplitude Modulation (AM) and Frequency Modulation (FM) halftone screening and the use of Cyan, Magenta, Yellow, and Black (CMYK) process color inks are endemic in commercial printing of all types, from high-volume offset lithographic printing to inkjet and toner printing in office and home environments. The pervasiveness of these graphic arts technologies means that criminals have ready access to them for counterfeiting. Just as security document substrates and security features are selected to be as different as possible from commercially-available materials, security document artwork can also be designed to resist counterfeiting. Security document design is often based on different artwork strategies that avoid the use of dots, and avoid the use of CMYK process color.

Using a novel shape or pattern other than dots is not the only strategy needed to create a secure document. Modern design software has made it incredibly simple to repeat a chosen shape that can be used as a halftone pattern. More secure documents use complicated patterns with change shape and size across the document, sometimes in a random and unpredictable way. These documents make it impossible to sample any particular area of a document to copy to another.

Proper security design will not only protect from counterfeiting by document re-origination but also by digital or photographic reproduction. There are many practices that assist in these goals. Security printers use particular colors that fall outside of the scanner and digital printer gamut to make copying difficult. Color choice as it applies to plate interaction is also important. Similar colors used on different printing plates across the same document, especially when used in combination with split fountain printing, can confuse counterfeiters when attempts are made to recreate a set of plates.

It takes a combination of skilled designers creating complicated security patterns with experienced printers utilizing secure inks and plate interactions to make a document that is difficult to counterfeit.

Security Printing, Documents, Document Design

J18 Analysis Techniques of Plastic Identity Documents

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After attending this presentation, attendees will gain a better understanding of new trends in plastic Identity Documents (IDs) and analysis methods by which to visualize and classify the materials used in their production.

This presentation will impact the forensic science community by increasing awareness of plastic identity documents and non-destructive/destructive analysis testing methodologies as a means of counterfeit deterrence and adversarial analysis, as well as identifying and highlighting tamper-resistant and copy-proof security features and printing incorporated within contemporary plastic IDs.

Identity documents are produced for and used by people around the world for a variety of different purposes, including proof of identity, employment eligibility, evidence of driving privileges, and domestic and international travel. Identity documents function as the link between an individual's government-verified biographical data (e.g., date of birth, nationality, and sex) and the individual himself. These documents must be inherently secure and tamper evident to maintain public confidence and trust.

Identity documents can be manufactured in many different ways with a variety of types of plastic, including polycarbonate, Poly-Vinyl Chloride (PVC), Polyethylene Terephthalate (PET), or a combination of these and others. These plastics are combined with heat, pressure, and/or adhesives to create an overall card body that guard against tampering or alteration. In addition, security inks, Optically Variable Devices (OVDs), and Radio Frequency Identification Devices (RFIDs) are incorporated within plastic documents as a deterrent against counterfeiting because they are difficult to replicate successfully. In addition, commercial availability of similar products and the knowledge of translating these products into counterfeit IDs are imperative for the manufacturer and designer to contemplate before placing a security feature into a document. Security printing is another important consideration in the manufacture of plastic identity cards; this includes personalization techniques such as laser engraving that discourages counterfeiters from replicating or compromising these methods.

Sophisticated non-destructive and destructive techniques that may be utilized to determine whether or not an ID is genuine will be explored. Non-destructive techniques including digital microscopy, which may be used to visualize plastic identity cards' security features from a three-dimensional, depth-up perspective, measure the relative heights of these features, and even classify the internal card body structure. Fourier Transform Infrared Spectroscopy (FTIR) is another non-destructive technique that allows the chemical composition of a variety of plastic substrates to be elucidated quickly and efficiently. A destructive technique that has been utilized at the laboratory in counterfeit deterrence analysis is card cross sectioning, which is achieved by cutting the card at a predetermined point of interest, mounting the card in an epoxy-resin mixture, polishing, and analyzing it with the Scanning Electron Microscope (SEM). Elemental techniques such as Energy-Dispersive X-Ray Spectroscopy (EDS) may then be employed to probe specific areas within the card body, such as the RFID, to ascertain an accurate elemental composition that was used in the card body construction.

Plastic, Identity Documents, Counterfeit Deterrence

J19 A Crosscut Shredded Document Case Made Easier: Part II — Predicting Where the Debit Card Pieces Go

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After attending this presentation, attendees will learn techniques that can facilitate the manual re-assembly of shredded debit, credit, or other rigid plastic cards.

This presentation will impact the forensic science community by proposing a methodology to make the process of manual re-assembly of shredded debit, credit, or other rigid plastic cards easier.

At the 2013 American Academy of Forensic Sciences meeting, a case involving a small bag of crosscut shredded paper documents was described. The case request was to reassemble them in the hopes of providing evidence in a case of identity

theft and the filing of false tax returns. Little could be found in the literature outlining procedures for reassembling shredded documents, but a methodology was eventually developed. One of the advantages of the methodology was the possibility of predicting the precise pattern into which a document was shredded.

The methodology includes, in part, the following steps:

- Sorting paper shreds by color and type of paper, and by markings present.
- Making measurements or comparisons of the width and length of the shredded pieces, and the angles formed by the machine-cut edges and the shredded edges.
- Orienting the pieces in the direction the document was shredded.
- Creating a template for assembly and a grid for aligning the shreds.
- Creating a grid of the pattern of the shred over the entire document.

Another shredded document case was subsequently received, this time involving reloadable debit cards. The cards were believed to have once been loaded with refunds obtained illegally through the filing of false tax returns, and the signature blocks on the back were assumed to bear the names of the return "filers," many of whose identities had been stolen.

The physical differences encountered between plastic shred and paper shred required some modifications in the methodology used for reassembling paper documents. Some of the steps modified involved how to:

- Prepare the shreds for handling.
- Straighten bent fragments.
- Estimate how many total cards were present.
- Affix the cards to a suitable medium for reassembling.
 - It also proved to be more a challenging process to:
- Align the pieces of the card being assembled.
- Predict the pattern into which each card was shredded.
- Despite the challenges, through the use of this revised methodology, parts of 45 cards were able to be assembled, providing the submitter with additional names and numbers important to the investigation.

Shredded Documents, Document Reconstruction, Plastic Card

J20 Revealing Writing That Has Been Covered Using Correction Tools

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After attending this presentation, attendees will learn of a novel method for revealing the underlying writing and/or printed material obliterated by various correction tools. In addition, this novel method of using the Electrostatic Detection Apparatus (ESDA) was compared to the use of the Video Spectral Comparator (VSC) for the same purpose. Attendees will also learn the benefits of using a non-destructive technique and the value of the VSC for this type of examination.

This presentation will impact the forensic science and questioned documents community by demonstrating the use of a common technique on a challenging type of evidence in an efficient manner.

One particular challenge encountered by the forensic questioned document examiner is that of obliterations through the use of correction utensils and opaquing solutions (e.g., Wite-Out®).

Underlying writing can be difficult to distinguish due to this type of obliteration. In the past, various methods have been implemented to deal with these types of evidence, such as physically removing the solution; however, this method has the potential to destroy the original evidence in the process.

An alternate non-destructive method for the visualization of the underlying writing might be preferred. The ESDA and the VSC are both non-destructive techniques widely used in the analysis of questioned documents. The ESDA was proposed and examined as a novel method for visualizing this type of obliterated writing. This non-destructive method was then compared to that of using the VSC for this same type of examination.

The evaluation was carried out through the use of a variety of writing instruments, inks, and printing methods. A series of samples consisting of handwritten, typewritten, and non-impact printed material was prepared and sections of each were covered with a different correction tool. Both the ESDA and VSC were used to view the underlying writing. The ESDA was able to distinguish the underlying writing in three of the eight samples where the correction tape was used. Surprisingly, for all other correction tools, the ESDA did not succeed in revealing the obliterated writing. In all cases, however, the VSC was able to generate a legible image of the original writing. In short, the VSC was exceptionally effective at visualizing writing obliterated by opaquing solutions. There may, however, be some situations where the ESDA may be advantageous.

ESDA, VSC, Correction Tools

J21 Analysis of Paper Textures Using High-Resolution Digital Photography and the Two-Dimensional Discrete Fourier Transform

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After attending this presentation, attendees will understand the application of the two-dimensional discrete Fourier transform to the discrimination of sheets of paper from different reams of paper.

This presentation will impact the forensic science community by demonstrating that two-dimensional discrete Fourier transform patterns can be obtained from sheets of paper using readily available digital camera equipment and ImageJ, an open source computer program.

Forensic document examiners may be asked to determine whether the paper on which a document was printed could have come from a particular source, such as a particular ream of paper. Such a problem can be approached using a variety of different measurements (e.g., thickness, color, fluorescence, Fourier transform infrared spectrometry, and X-Ray diffraction). Ideally, forensic methods of analysis for paper should be non-destructive and inexpensive. Paper manufacture can introduce repetitive patterns to the surface of the paper. These can be visualized using transmitted light, a procedure often used to visualize and photograph watermarks. The two-dimensional discrete Fourier transforms of transmitted light images of sheets of paper provide a way of visualizing repetitive patterns and of more objectively comparing them. The two-dimensional discrete Fourier transform produces images with patterns of spots which reflect the periodicity of repetitive patterns and their directionality. The research reported here explored whether forensically useful two-dimensional Fourier transform papers could be obtained using simple photographic

equipment and open source computer software.

Sheets of white office paper were selected from each of 16 reams of paper. These reams of paper came from nine different vendors. Each sheet was placed on a white light transilluminator and three to five images of different areas on the sheets were captured with a 14-megapixel digital single lens reflex camera mounted on a tripod. It was found that exclusion of stray light and sharp focusing of the camera were critical to obtaining usable digital images. Two-dimensional discrete Fourier transforms were computed for each digital image using ImageJ, an open source computer program developed by the National Institute of Health for image analysis. Versions of this program are available for different computer operating systems. The images were converted to gray scale and cropped to provide a square image. This was done because the two-dimensional discrete Fourier transform algorithm employed by ImageJ requires square images. To allow the Fourier transform patterns to be more easily viewed, the image produced by the two-dimensional Fourier transform was converted to binary (black and white) and eroded twice to remove noise from the image.

The Fourier transform patterns obtained from each sheet proved to be highly reproducible. Each sheet of paper was found to produce a unique Fourier transform pattern. Moreover, these patterns were different from those reported by researchers at the Netherlands Forensic Institute. Further research must be done to determine whether the Fourier transform patterns are consistent within a ream of paper.

This presentation demonstrates that two-dimensional discrete Fourier transform patterns can be obtained from sheets of paper using readily available digital camera equipment and ImageJ, an open source computer program.

Forensic Document Examination, Paper, Fourier Transform

J22 Characterization and Discrimination of Printing Inks Using DART®-MS, Py-GC/MS, and ATR-FTIR for Forensic Document Analysis

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Attendees will learn that Direct Analysis in Real Time Mass Spectrometry (DART®-MS), Pyrolysis Gas Chromatography Mass Spectrometry (Py-GC/MS), and Attenuated Total Reflectance-Fourier Transform Infrared (ATR-FTIR) Spectroscopy are viable methods through which inkjet inks and toners can be characterized in order to distinguish between inks and toners from different sources and associate inks and toners printed from the same source.

This presentation will impact the forensic science community by demonstrating how the combination of these three techniques provides information about the chemical profile of an ink or toner that can be used in forensic document analysis.

A total of 150 inkjet and toner samples (75 of each type) were analyzed via the three analytical methods. Each sample consisted of a monochromatic Cyan, Magenta, Yellow, and Black (CMYK) inkjet or toner printed or manually deposited onto Whatman® 42 paper. It was determined there is no significant difference in chemical profiles obtained between printed and manually deposited inkjets or toners on paper.

For ATR-FTIR, analysis was performed using a Perkin® Elmer® Spectrum One FTIR with Pike Technologies GladiATR™ Diamond Single Bounce Crystal accessory with imaging capabilities.

Samples were analyzed directly on paper without destruction. Samples could be analyzed in under a minute each. Based on the chemical information obtained for toners, the 75 samples were divided into 12 different groups based on polymer content observed in the IR spectra. Toners could be easily separated into groups based on the presence or lack thereof of polystyrene, acrylate, epoxy, and polyethylene resins. However, no chemical information was obtained for inkjet samples due to strong paper background contribution.

For Py-GC/MS, an Agilent Technologies® 7890A GC with 5975 inert XL MSD, and CDS Analytical 1500 series pyroprobe were used for analysis. Toner samples were transferred to aluminum foil for analysis to remove interference of paper background at high temperature. The chemical information obtained for toners included different resins present such as toluene, styrene, substituted benzene, and naphthalene that allow for classification. Inkjets were analyzed at a lower pyrolysis temperature to desorb volatile components from the inks on paper. This method does destroy a 1x10mm cut of ink on paper. For inkjets, volatile organic compounds such as diols, pyrrolidinone, glycols, and glycerols were observed that make up the chemical profile.

For DART®-MS, analysis was performed using an Agilent 6530 Q-TOF mass spectrometer and IonSense DART®-SVP. Both toner and inkjet samples were analyzed directly on paper with no sample preparation or destruction. A spectrum for each sample could be produced in less than two minutes. The mass spectra for inkjets include Polyethylene Glycol (PEG) mass fragments with differences of 44 Da that are characteristic for each sample. Toner samples also produce characteristic spectra with polymer-like distributions that are significantly different from the paper background.

Overall, the combination of these techniques and the fusion of the data provides comprehensive information to distinguish between inks and toners printed from different sources and associate inks and toners printed from the same source without requiring extensive sample preparation or total destruction to a document.

Inkjets, Toners, Spectrometry



Toxicology



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K1 Patterns of Drug Use in a Suicide Population by LC-TOF Analysis of Whole Blood Samples for Common Therapeutic, Abused, and Novel Psychoactive Drugs

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After attending this presentation, attendees will be able to apply knowledge about demographics and patterns of drug use related to cause and manner of death in a cohort of subjects who successfully committed suicide by a variety of means. Additionally, attendees will be able to assess the value of performing drug screening in suicide cases that are not apparent drug overdoses.

This presentation will impact the forensic science community by increasing the knowledge about patterns of drug use in a suicide population including the presence or absence of recently emerging novel psychoactive substances ("Bath Salts").

Due to the increase in use, legal complications, and potentially dangerous effects of these designer drugs, obtaining a better understanding of their prevalence is an important insight for coroners and medical examiners. Currently, pathologists and coroners have highly variable policies when it comes to toxicology testing of cases deemed a suicide in which there is clear trauma (Gunshot Wounds (GSWs), hangings, blunt force injuries, etc.). Cases considered suicides will often include a partial autopsy but no toxicology screen. This is a result of a combination of a lack of resources together with a lack of understanding of the role played by prescription or abused drug use in suicidology. Helping develop this knowledge base will assist with the development of more uniform standards for death investigation. Over a 12-month period (2012-2013), blood samples were collected for sequential cases determined to be suicides (n=145). Apparent drug-related deaths (n=22) had previous toxicology studies performed, while the remainder (n=123) did not.

Using a Liquid Chromatography-Time Of Flight (LC/TOF) screening method, the 145 subject samples were screened with both a standard toxicology panel and a novel psychoactive-substances screening panel (stimulants) using LC/TOF methods. The method tested for 305 commonly encountered drugs and their metabolites in the categories, including drugs of abuse and therapeutics. The scope did not include tetrahydrocannabinol (THC). The samples were tested for alcohol by Headspace/Gas Chromatography/Mass Spectrometry (HS/GC/MS). Method validation was performed, followed by quantitation of the samples containing the synthetic stimulants. The results were examined for patterns and relative frequencies of drug use within different causes of death, race, sex, and age.

The subjects ranged from 13 to 95 years of age (mean=44.8 and median=44). Four of the subjects were minors under the age of 18 years with the youngest being 13 years old. In two of the four subjects under 18 years of age, no significant

drugs were found. Desmethylsertraline was found in the 13-year-old while acetaminophen, dihydrocodeine/hydrocodone, ephedrine/pseudoephedrine, fluoxetine, hydrocodone, lamotrigine, lorazepam, methylphenidate, trazodone, and ziprasidone were found in the 16-year-old. The majority of subjects were listed as Caucasian (80.7%). Based on a 2012 estimate, 67.1% of the population in Marion County is Caucasian (includes persons reporting only one race).

The drugs detected were categorized into 17 groups: Alzheimer's disease medication; anticoagulant; anticonvulsant; antidepressant; antihistamine; antipsychotic; benzodiazepine; cardiac drug; Erectile Dysfunction (ED) drug; hallucinogen; muscle relaxant; narcotic analgesic; opioid antagonist; sedative; sleep medication; Selective Serotonin Reuptake Inhibitor (SSRI); and stimulant. The categories with the highest prevalence were narcotic analgesics (28.3%: dihydrocodeine/hydrocodone (n=16); oxycodone (n=10); and fentanyl (n=8)), benzodiazepines (22.8%: diazepam (n=9); alprazolam (n=8); and lorazepam (n=7)), and stimulants (19.3%: amphetamine (n=7); hydroxybupropion (n=7); and cocaine (n=6)).

The causes of death were broken down into six categories: GSW (49.0%); asphyxia (24.8%); drug intoxication/Overdose (OD)/toxic chemicals (16.6%); Carbon Monoxide (CO) inhalation/fire death (4.8%); sharp force injury (2.8%); and blunt force injury (2.1%). There were a significantly higher number of male subjects (n=117, 80.7%) than females (n= 28, 19.3%). Within the GSW deaths, the most frequently encountered drugs were oxycodone (n=7), hydrocodone (n=7), and diazepam (n=6). GSW was the most prevalent of groups with 71 cases, 59.2% of which contained drugs. Asphyxia deaths were the second most common with a total of 36 cases, with 58.3% containing drugs. Within the deaths by asphyxia, the most frequently encountered drugs were amphetamine (n=5) and hydroxybupropion (n=3). Drug intoxication/OD/toxic chemicals made up 24 cases, with 95.8% of the cases containing drugs. The most prevalent drugs in this category were alprazolam (n=6), dihydrocodeine/hydrocodone (n=6), and hydrocodone (n=6).

Synthetic Stimulants, Suicide, Drug Overdose

K2 Method Development for the Combined Analysis of Synthetic Cannabinoids and Designer Cathinones in Urine Specimens

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After attending this presentation, attendees will have an understanding of a combined high-throughput analytical screening method for the detection of both designer cathinones and synthetic cannabinoids in urine by Liquid Chromatography Tandem Mass Spectrometry (LC/MS/MS).

This presentation will impact the forensic science

community by providing a high-throughput method for the detection of emerging drugs of abuse.

Since their emergence in 2009, designer drugs continue to gain popularity on the recreational drug use market. Synthetic cannabinoids and substituted cathinones are marketed to drug users as legal alternatives to marijuana and methamphetamines, respectively. In addition to easy access and affordability, the endless changes to chemical composition enable drug users to avoid detection in standardized drug tests while posing significant challenges to forensic scientists. As a means of deterrence for its service members, the United States Navy has recently instituted a testing laboratory capable of detecting several of these compounds. Individual validated extraction and detection methods are used for each drug class, which are both costly and time consuming. This will become progressively more evident as the required number of service member specimens increases.

The purpose of this study was to modify and combine the two existing methods and develop a single high-throughput urinary drug screen sufficient to detect both synthetic cannabinoids and designer cathinones. Six synthetic cannabinoids (JWH 073 N-5-COOH, JWH 018 N-5-COOH, AM2201 N-4-OH, MAM 2201 N-5-COOH, MAM 2201 N-4-OH, and UR 144 N-5-COOH) and seven cathinones (methylenedioxypyrovalerone (MDPV), 4-fluoroamphetamine (4-FA), mephedrone, 4-methylethcathinone (4-MEC), α -pyrrolidinovalerophenone (α -PVP), methylone, and butylone) were used in the method development with Liquid-Liquid Extraction (LLE) and detection by LC/MS/MS using electrospray ionization in multiple reaction monitoring mode. Data from this study details a high-throughput screening method that is validated for repeatability, precision, and accuracy. The new synthetic cannabinoid and cathinone screening method will provide a more efficient means of testing specimens that is in alignment with the overall mission.

Cathinones, Synthetic Cannabinoids, Method Development

K3 Detection of Inhalants and N-Propanol in Forensic Toxicology: A Six-Month Review

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After attending this presentation, attendees will understand the procedure for identifying inhalants and Volatile Organic Compounds (VOCs) using headspace sampling coupled with Gas Chromatography-Mass Spectrometry (Headspace GC/MS), the prevalence of inhalants in biological specimens, and the potential risk of using the internal standard, n-propanol, in postmortem sample analyses.

This presentation will impact the forensic science community by offering insight to the qualitative analysis of common inhalants observed in biological samples. This study also discusses the use of n-propanol as an internal standard for postmortem analyses.

Due to their extensive availability, low cost, and potential for euphoric effects, inhalants have the potential for abuse. They are present in many commercial products including paint thinners, nail polish removers, degreasers, dry-cleaning fluids, correction fluids, and various aerosol products. The main objective of this study was to determine the prevalence of inhalant use through the analyses of blood or similar biological specimens. Chemicals of interest include, but are not limited to, 1,1-difluoroethane, difluorochloromethane, 1,1,1,2-tetrafluoroethane, nitrous oxide, propane, methanol, and toluene. A secondary element of this study

was to look at the prevalence of n-propanol in postmortem cases as a postmortem artifact. The complex processes of decomposition produce a variety of chemicals as the soft tissues break down and n-propanol may be generated as a by-product. N-propanol is often used as an internal standard in the quantitation of ethanol and similar volatile compounds.

Headspace GC/MS is a useful method of analysis for VOCs because it requires minimal sample preparation. Six months of postmortem and Driving Under the Influence (DUI) case samples were analyzed to determine the prevalence of inhalants and n-propanol in biological samples. An Agilent® 7890A gas chromatograph equipped with a 7697A Headspace Autosampler and Agilent® 5975C mass spectrometer was utilized for this study. The GC capillary column used was a DB-1 column 30m x 0.32mm x 5.0 μ m.

Of 783 cases analyzed, four cases returned positive results for inhalants. 1,1-difluoroethane was present in three of the cases. 1,1-difluoroethane is a halogenated aliphatic compound that is a colorless and odorless gas at standard temperature and pressure. It is the main component of some gas dusters and dust removers used to clean electronic devices. It can cause nausea, vomiting, confusion, dizziness, drowsiness, lethargy, depressed reflexes, muscle weakness, stupor, and, in extreme cases, coma or death.

Difluorochloromethane (chlorodifluoromethane, Freon® 22), an air conditioner fluid, was detected in one sample. Difluorochloromethane is a halogenated hydrocarbon that, when inhaled, can cause symptoms similar to other inhaled gases. All four cases positive for inhalants were postmortem cases where heart blood was tested. Of the 557 postmortem specimens analyzed, n-propanol was observed in 25 cases; 19 were chest fluids (cases in advanced stage of decomposition), the other 6 were heart bloods. Of the 41 chest fluids analyzed, 19 (43%) were positive for n-propanol. Other cases showed indications of n-propanol; however, the abundance was insufficient for identification. It was concluded that the prevalence of n-propanol in postmortem samples was not significant enough to warrant a change of internal standard; however, in cases of decomposition, it should be taken into consideration when evaluating samples.

Inhalants, Headspace GC/MS, N-Propanol

K4 Detection of 18 Synthetic Cathinones and New Stimulants in Urine

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After attending this presentation, attendees will gain knowledge on the performance of a new analytical method developed in order to detect a wide range of new psychoactive substances in urine.

This presentation will impact the forensic community by describing which molecules among new synthetic cathinones or other new stimulants are likely to be the most used in the Piedmont Region of Northern Italy.

Recently, forensic laboratories have been challenged worldwide with the need to detect a variety of New Psychoactive Substances (NPSs) in different biological specimens. These compounds, frequently referred to as either "designer drugs" or "legal highs," have little or no previous history of medicinal use. Although the consumption of these drugs has been tolerated for quite

some time, currently the majority of them are progressively banned as serious side effects and acute intoxications (occasionally fatal) are becoming increasingly frequent. However, legal interdiction of specific substances is often thwarted because their chemical structure can be slightly modified with relative ease to create new, still-unbanned drugs with similar properties. Amphetamine derivatives, with backbone molecular structures of either piperazine or cathinone, represented the initial group of designer drugs. Although most of the latest drugs act as Central Nervous System (CNS) stimulants, their chemical structures are assorted, ranging from derivatives of pipradrol, ketamine, phencyclidine, arecoline, aminopropylbenzofuran, and ring-substituted aminoindans to thiophenyl bioisosteres of methamphetamine, as well as cocaine-related structures. Several analytical methods were developed to determine some NPSs and/or their metabolites in either blood or urine, but routine urine screening tests are rarely performed on large populations. Consequently, almost no prevalence studies are available. For the same reason, the potential association between NPS use and traffic or occupational accidents is totally unknown so far.

This study developed and validated a Gas Chromatography/Mass Spectrometry (GC/MS) method to detect 18 compounds in urine. The following parameters were investigated: selectivity; specificity; linearity range; detection and quantification limits (Limit of Detection (LOD) and Limit of Quantitation (LOQ)); intra-assay and inter-assay precision and accuracy; carry-over effect; recovery; and matrix effects. Finally, the method was applied to the analysis of 43 real urine samples, obtained from roadside drug controls and workplace drug testing. Four samples tested positive for butylone, one for mephedrone, and nine for cathine/ephedrine, including one sample also positive for 4-fluoroamphetamine.

This study demonstrated the spread of new synthetic cathinones or other new stimulants, likely as recreational drugs and potentially used as substitutes for cocaine and amphetamines, in the population of drug consumers in Piedmont, Northern Italy. Therefore, the progressive introduction of efficient screening and confirmation tests for the detection of acute and chronic abuse of the new "designer-drugs" appears to be crucial within the entire drug-prevention policy.

Cathinones, Bath Salts, Mephedrone

K5 Quantitative Analysis of 11-Nor-9-Carboxy-Tetrahydrocannabinol in Hair by Column Switching LC/ESI/MS3

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After attending this presentation, attendees will better understand the column switching technique of High-Performance Liquid Chromatography (HPLC) as well as Liquid Chromatography/Electrospray Ionization/Tandem Mass Spectrometry³ (LC/ESI/MS³) to decrease background hair matrix to identify and quantify 11-nor-9-carboxytetrahydrocannabinol (THCCOOH), a metabolite of tetrahydrocannabinol (THC) in hair samples.

This presentation will impact the forensic science community by providing a new sensitive and selective method of hair analysis by the column-switching technique of HPLC and LC/ESI/MS³. This method can be used as an alternative method for

Gas Chromatography-Mass Spectrometry (GC/MS) analysis of cannabis in hair without derivatization.

Hair analysis has been regarded as an alternative method for urine analysis in forensic and criminal cases. Cannabis (marijuana) is one of the most widely used drugs in the world and it has been controlled in South Korea since 1976. Identification of THCCOOH in hair can be an important proof of cannabis use because it can exclude the possibility of passive cannabis smoke exposure. This study describes a quantitative method of THCCOOH in hair using simple Liquid-Liquid Extraction (LLE) and selective column-switching LC/ESI/MS³.

The column-switching valve was placed in the column compartment. A pre-column (C₁₈, 2.0 × 30mm, 3.0μm) was used in the modified gradient method. For the column-switching system, the trap column (C₁₈, 1.0 × 30mm, 2.6μm) and the analytical column (C₁₈, 2.1 × 75mm, 2.7μm) were used. The valve switch from precolumn to trap column was set from 3.0 to 4.0min. The sample extract was injected on the precolumn which was flushed with 2mM ammonium formate/0.2 % formic acid in water and 2mM ammonium formate/0.2 % formic acid in acetonitrile at a flow rate of 0.5ml/min. THCCOOH appeared around 3.5min in this precolumn. From 3.0 to 4.0min, the analytes were flushed onto the trap column with 0.5mM ammonium formate in water and 0.5mM ammonium formate in acetonitrile. After 4.0min, the valve was switched to the original position and the analytes in the trap column were eluted onto the analytical column. Resolution occurred in this column by increasing the ratio of organic solvent and finally eluted into the ESI/MS³ system. The internal standard was THCCOOH-d₃.

In the MS³ experiment, THCCOOH ionized best in negative ESI mode and an (M-H)⁻ ion was observed at *m/z* 343, which was fragmented into the second precursor ion at *m/z* 299.2. This second precursor ion was trapped and accumulated in the Linear Ion Trap (LIT) using a fixed LIT fill time of 250ms and excitation time of 20ms. The resulting MS³ spectrum showed an intense peak at *m/z* 245.1; therefore, for the quantification of THCCOOH, the MS³ ion transition monitored was *m/z* 343.2 → 299.1 → 245.2 (343.1/299.2/245.1) with the LIT set to perform a mass scan centered at *m/z* 245.1. For the internal standard (THCCOOH-d₃), the MS³ ion transition monitored was 346.2 → 302.2 → 246.1 (346.1/302.2/246.1) in the same method.

Chromatographic separation was completed within 12min. No interferences were detected in 10 blank hair samples. The correlation coefficients (*r*²) of calibration curves were larger than 0.9997 with mean slope of 0.0202 and the mean intercept of 0.0017, using a weighing factor 1/*x*. In the intra- and inter-assay precision and accuracy study, Coefficient of Variation (CV) (%) and bias (%) were below 10. The limit of detection was 0.08pg/mg and the limit of quantification was 0.1pg/mg. The mean values of matrix effect at 10pg/20mg and 50pg/20mg were 82.8 and 70.6%, respectively. The CVs of the matrix effect, a measure of the relative matrix effect for an analyte, at each concentration were 9.5 and 6.1%, respectively, which showed no significant variation among hair samples from different individuals. The mean values of recovery were 91.7 and 74.0% and those of process efficiency were 80.6 and 66.3% at 10 and 50pg/20mg hair, respectively.

The range of concentrations of THCCOOH from 94 authentic human hair samples was 0.1 ~ 15.7pg/mg. This method was successfully applied in the analysis of authentic human hair samples. The developed method will be very useful for quantification of THCCOOH in hair in both legal and public health aspects.

Cannabis, Hair, LC/MS/MS

K6 Benzodiazepine Effects on Opioid Parent

and Parent-to-Metabolite Concentrations

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After attending this presentation, attendees will be able to characterize the parent drug to metabolite ratios for fentanyl, hydrocodone, methadone, and oxycodone in the presence and absence of benzodiazepines and note potential differential effects of alprazolam and diazepam on blood opioid concentrations in unintentional deaths.

This presentation will impact the forensic science community by contributing to the understanding of the possible interactions between benzodiazepines and opioids in unintentional drug deaths and assisting in the interpretation of parent opioid and metabolite concentrations during death examinations.

Drug overdose deaths involving opioids are a major health concern. They typically involve multiple drugs, with benzodiazepines (usually diazepam or alprazolam) frequently present. Many toxicology laboratories routinely measure parent drug and metabolite concentrations in poisoning deaths. The parent drug to metabolite ratio can help determine whether a drug overdose was acute, the manner of death, abuse/misuse vs. therapeutic drug use, or to differentiate between ingestion of a drug vs. an active metabolite that is also a prescribed drug. Few studies have examined postmortem blood opioid concentrations and parent drug/metabolite ratios in combination opioid-benzodiazepine deaths. Such studies can better characterize the range of values found and whether the opioid parent drug/metabolite ratio might be affected by benzodiazepines.

A Forensic Drug Database (FDD) was initially created to capture West Virginia (WV) drug death data. A project funded by the WV Injury Control Research Center's Centers for Disease Control and Prevention (CDC) renewal grant will expand the FDD to the Northern New England (NNE) states. Decedent data entered into the FDD includes demographic information, body condition, Body Mass Index (BMI), death certificate data, route of drug administration, whether a prescription was present for controlled substances identified, medical history, key autopsy findings (all decedents), and toxicological analyses (for all drug-related deaths). The database currently contains information on 2,784 WV drug-related deaths from January 2005 through most of 2010 (data entry ongoing); data from NNE are being compiled.

This study evaluated postmortem peripheral blood concentrations of parent drug and parent/metabolite concentration ratios of four opioids that are at least partially metabolized by Cytochrome P450 3A4 (CYP3A4): fentanyl/norfentanyl; hydrocodone/dihydrocodeine (metabolite most commonly measured); methadone/EDDP; and oxycodone/oxymorphone, in the presence and absence of co-intoxicant benzodiazepines, alprazolam, and diazepam. Due to potential competition for, or inhibition of, CYP3A4 by alprazolam or diazepam, the opioid parent drug concentrations and parent-to-metabolite ratios might be expected to be higher in the presence of these benzodiazepines compared to their absence.

All accidental WV overdoses involving ≤ 4 concomitant drugs (to reduce possible confounding) and in which fentanyl, hydrocodone, methadone, or oxycodone caused or contributed

to death were identified. Cases were excluded if >1 opioid was present or if benzodiazepines other than alprazolam or diazepam were found (Dataset A: fentanyl (n=135); hydrocodone (n=135); methadone (n=337); oxycodone (n=270)). For parent drug and concentration ratio analyses in the presence/absence of benzodiazepines, cases with co-intoxicants that were inhibitors or inducers of CYP3A4 or Cytochrome P450 2D6 (CYP2D6) or that had documented pharmacokinetic interactions with opioids were further excluded (Dataset B: fentanyl (n=56); hydrocodone (n=49); methadone (n=166); oxycodone (n=78)).

Median concentrations were used for analyses. A summary of key findings follows. As the number of concomitant drugs increased, hydrocodone, methadone, and oxycodone concentrations significantly decreased. Statistically significant reductions in fentanyl and methadone concentrations (42% and 44%, respectively) were found when decedents lacked a valid prescription for that drug compared to when one was present (Dataset A). Only hydrocodone parent concentrations were significantly different overall in the presence or absence of benzodiazepines. Co-ingestion of alprazolam or alprazolam + diazepam were likely responsible (reductions of 34% and 47.5%, respectively). Only the methadone parent/metabolite ratio was significantly affected by benzodiazepines, with a 35% reduction with diazepam (11.8 vs. 7.7) (Dataset B).

In conclusion, lower concentrations for certain opioids were found in deaths with increasing numbers of co-intoxicants and when a valid prescription was absent. Opioid metabolism is a complex process, affected by a number of factors. Some differential effects were present in parent and metabolite concentrations among the opioids studied with and without co-ingested benzodiazepines. Further studies are needed to characterize the factors that individually or when combined might affect opioid concentrations in unintentional overdose deaths.

Opioids, Benzodiazepines, Unintentional Overdoses

K7 A Validated Analytical Method for Simultaneous Detection of ATS in Human Urine Using SPME-GC/MS

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After attending this presentation, attendees will understand the benefits of using Solid-Phase Microextraction-Gas Chromatographic/Mass Spectrometry (SPME-GC/MS) to analyze Amphetamine-Type Stimulants (ATS) in human urine and the quantification results of authentic urine samples.

This presentation will impact the forensic science community by presenting a validated analytical method for simultaneous detection of ATS in human urine.

The Fluorescence Polarization Immunoassay (FPIA) as a preliminary test sometimes gives false positive results due to structural similarity between ATS. A rapid and easy SPME-GC/MS screening method has been developed and validated for simultaneous detection of nine ATS in human urine. These drugs include methamphetamine, amphetamine, Methylenedioxymethamphetamine (MDMA), and 3,4-methylenedioxyamphetamine (MDA) which are popular abused drugs in Korea. Phentermine, phenmetrazine, ephedrine,

pseudoephedrine, and norephedrine/norpseudoephedrine also show cross-reactivity by amphetamine FPIA.

The conditions of SPME extraction time, temperature, and deposition time were optimized to yield the highest peak area. The extraction condition using 100µm Polydimethylsiloxane (PDMS) fiber was 80°C for 25min with 250rpm and deposition time was 4min in the GC injector at 280°C with split mode (ratio 5:1). To prevent carryover, the fiber was baked out pre- and post-injection into the GC/MS. The GC separation was performed using temperature program of 100-300°C at 10°C/min.

The data was collected by extracted ion chromatogram for each drug from total ion chromatograms by full scan mode and calculated by peak area ratios (peak area drug/peak area internal standard). All nine ATS showed good resolution with retention times from 4.45min to 9.33min. Methamphetamine and ephedrine were especially easy to discriminate by retention time. High concentrations of ephedrine and low concentrations of methamphetamine in urine are difficult to distinguish from each other because of similar retention time and similar fragmented ions by Pentafluoropropionic Acid Anhydride (PFP) derivatization. The calibration curve showed acceptable linearity for each drug with $R^2 > 0.99$. The results of the intra- and inter-day precision and accuracy were satisfactory: <10% for precision and within $\pm 10\%$ for accuracy at three different concentrations (167, 333, 1,000ng/mL). In this analytical method, no significant matrix effect was observed and high recoveries (>92%) were achieved. Thus, SPME-GC/MS has the advantages of an easy sample preparation with acceptable accuracy and precision for the simultaneous quantification. It will be a useful method for a simple and rapid screening analysis of urines for ATS abuse.

SPME-GC/MS, Amphetamine Type Stimulants, Validation

K8 Short-Term Stability of Mephedrone in Blood and the Impact of Storage Conditions on Concentrations Detected by GC/MS

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After attending this presentation, attendees will understand the implications of different storage conditions upon mephedrone concentrations in blood.

This presentation will impact the forensic science community by offering forensic toxicologists, chemists involved with drug analysis, and forensic pathologists more information about appropriate sample storage conditions to reduce potential sample degradation and loss between the time samples arrive into the laboratory and when they are tested.

Introduction: The time between sample collection and sample analysis varies greatly from case to case; therefore, knowledge of analyte stability is of extreme importance. Not only does it help the toxicologist select the most suitable sample for analysis, it also ensures that optimum storage conditions and preservatives are used to limit any sample degradation. This in turn aids the interpretation of concentrations of any drugs detected and their significance.

Mephedrone first appeared on sale to the public in 2007, and remains one of the most routinely detected "bath salts" or Novel Psychoactive Substances (NPS) in the United Kingdom and the United States. At the time, the majority of toxicology laboratories were not equipped to test for this compound; however, since developing methods for its analysis, many have performed retrospective testing on samples to investigate the presence of this drug. Previous work has shown mephedrone to be unstable

in biological matrices. This research was intended to ascertain the effects of various storage temperatures and preservatives in preventing analyte degradation and determine the optimum conditions.

Method: Preservative-free bovine blood was spiked with mephedrone (1mg/L). This was divided into separate aliquots (1mL), enabling the examination of the stability of the drug in blood stored: (1) without preservative; (2) with citric acid preservative (8%); and, (3) with sodium fluoride (1.67%)/potassium oxalate (0.20%) at three different temperatures (-20°C, -4°C, and 20°C). Aliquots were tested daily for seven days and then weekly over a period of ten weeks. Samples were analyzed in triplicate and calibrations and controls were run during each analysis. Samples were extracted using Solid Phase Extraction (SPE) prior to derivatization with Pentafluoropropionic Acid-Ethyl Acetate (PFP:EtOAc (2:1)) and analyzed using a Bruker® Gas Chromatograph-Tandem Mass Spectrometer (GC/MS/MS) with a DB5 column (30m x 0.25mm, 0.25µm) in splitless mode. The total run time was 25 minutes, and mephedrone-D₃ was used as the internal standard.

Results: Samples stored at 20°C were the most effected with mephedrone becoming undetectable after a period of 21 days regardless of any preservative present. After one day, samples stored at 20°C saw a loss of on average 19%, ranging from 17% with citrate solution to 21% with fluoride oxalate. Refrigerated samples preserved with fluoride/oxalate and citrate preservatives were initially stable; however, fluoride/oxalate rapidly decreased after five weeks, with a total loss of 96% over the ten-week period. Refrigerated samples preserved with citrate solution showed no significant decrease over the ten-week period. Refrigerated samples stored with no preservative saw a 41% drop after five weeks and a 74% reduction in concentrations across the ten-week period. Samples stored at -20°C were stable under all conditions over the ten-week period.

Conclusion: To maximize stability of mephedrone, samples should be stored at -20°C and preserved using citrate as this prevents any oxidative losses occurring. Although fluoride/oxalate was shown to preserve samples when stored at -20°C, degradation was still problematic at 4°C after five weeks, seeing a 36% decrease. This in turn would affect reported concentrations from laboratories which store samples at 4°C prior to analysis before archiving them at -20°C. Regarding retrospective analysis, analysts should be cautious in interpreting negative results in cases where the history indicates mephedrone use. The sample history must be taken into consideration. Further work is underway to investigate potential degradation products; cross validation with human blood is also being planned.

Stability, Mephedrone, Preservatives

K9 Gummy Bear Candies Containing Designer Drugs in a Drug Facilitated Sexual Assault (DFSA) Case

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After attending this presentation, attendees will gain insight into the use of designer drugs in the commission of a Drug Facilitated Sexual Assault (DFSA) and the manufacture of candies as the means of delivery.

This presentation will impact the forensic community by highlighting a unique drug dosage form and the complexity of a

DFSA investigation.

A 21-year-old female residing in Lake County, Ohio, reported a sexual assault by three males (22, 24, and 26-years-old) whom she had just met. The males, from the New York City area attending welding school in Cleveland, Ohio, arranged to meet the female at her residence. One male offered the female a "gummy bear" candy from a large bag containing numerous gummy bear-shaped candies, stating it was an "upper" bear. The female admitted to willfully ingesting the gummy bear and shortly thereafter feeling numb and unable to move her arms.

Another male then lifted the female's head and repeatedly forced her to inhale an unknown white powder. The female conveyed she felt "paralyzed" prior to the sexual assault by the males. The female filed a police report 23 days later and provided one gummy bear and one glass vial containing residue that had been left at her residence after the assault. The female contacted a local rape crisis center, but a rape kit was not collected due to the delay in reporting. No toxicology samples were collected.

Analysis of the gummy bear revealed the presence of 6-(2-aminopropyl)-2,3-dihydrobenzofuran (6-APDB), an entactogen analog of 3,4-methylenedioxyamphetamine (MDA). Analysis of the vial residue revealed the presence of methoxetamine (MXE), a dissociative anesthetic derivative of ketamine.

Approximately 12 hours after the assault, the three males were traveling in a vehicle that was stopped for speeding and a lane violation. The driver failed field sobriety tests and was arrested for operating a vehicle under the influence of alcohol or drugs (OVI). The front and rear passenger were observed to be lethargic with glassy eyes and slow speech. Both passengers subsequently passed out during an inventory of the vehicle.

Driver toxicology was negative for ethanol in whole blood and urine and screened positive for cannabinoids in whole blood. The driver admitted to drinking two beers and smoking marijuana earlier. The rear passenger stated he snorted MXE prior to the traffic stop. The front passenger stated he uses clonidine and naltrexone to "take the edge off" heavy withdrawal.

A backpack discovered during the vehicle inventory contained numerous purported drugs and drug paraphernalia including strainers, a liquid eye dropper, a syringe, spoons, scissors, tweezers, empty gelatin boxes, "bear" candy molds, a "tree" candy mold, a round shape candy mold, rolled paper, digital scales, a grinder, smoking pipes, plant material, plastic bags containing powders (white/yellow/brown), tablets (white/tan/green), and numerous gummy bear candies (red/yellow/orange-green).

Analysis of the backpack items revealed the presence of the following drugs: MXE; 6-APDB; 5-(2-aminopropyl) benzofuran (5-APB); 6-(2-aminopropyl)benzofuran (6-APB); 4-fluoroamphetamine (4-FA); fluoromethamphetamine (FMA); 4-chloro-2,5-dimethoxyamphetamine (DOC); 4-acetoxy-N,N-dimethyltryptamine (4-AcO-DMT); 2,5-dimethoxy-4-ethylphenethylamine (2C-E); 3,4-methylenedioxypropylone (MDPV); 3,4-methylenedioxyamphetamine (MDMA); phenazepam; methamphetamine; JWH-250; marijuana; naltrexone; and clonidine. The red gummy bear candies contained 6-APDB, the yellow gummy bear candies contained 4-FA, and the orange-green gummy bear candies contained phenazepam.

The female advised she was uncertain whether she would follow through with the DFSA investigation and ceased contact with the detective. The three males were sentenced to limited jail time, driver's license suspension, and community control. Additionally, the driver was required to pay a fine, complete a relapse prevention program, and report to adult probation.

Designer Drugs, DFSA, Analog

K10 Parameter Effects in a Hydrophilic-Interaction Liquid Chromatographic Method to Simultaneously Quantify Polar Metabolites and Closely Associated Parent Opiates in Urine

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After attending this presentation, attendees will have a better understanding of the capabilities of Hydrophilic-Interaction Liquid Chromatography (HILIC). HILIC has the ability to simultaneously retain polar glucuronide metabolites while still separating structural isomers and less polar parent drugs.

This presentation will impact the forensic science community by providing information of parameter changes in a HILIC Tandem Mass Spectrometry (MS/MS) method which can simultaneously quantify 6-monoacetylmorphine (6-MAM), morphine, hydrocodone, codeine, oxycodone, hydromorphone, oxymorphone, codeine-6-glucuronide, morphine-6-glucuronide (M6G), and morphine-3-glucuronide (M3G) in urine.

HILIC has become a chromatographic tool to better retain polar analytes where reverse phase chromatography cannot. HILIC uses a mixed mode system involving a polar stationary phase and a mostly organic mobile phase with some water. The polar water component of the mobile phase favors the hydrophilic moieties of the column packing and creates a stagnant layer in which a partition occurs with the organic component of the mobile phase. This allows for polar analytes to interact with the column more and elute later.

In this presentation, zwitterion-HILIC demonstrates the ability to analyze low concentrations of polar metabolites without sacrificing selectivity between the parent opiates. Zwitterion-HILIC consists of a specific stationary moiety that contains both amino and sulfate groups to allow for both positive and negative interaction with the analyte. Analytes chosen are morphine and codeine and their glucuronides, M6G, M3G and C6G. Although M3G and M6G are structural isomers, they have different pharmacological effects. When analyzed separately, M6G can provide intoxication levels and M3G can elucidate predisposition of abuse. Also hydromorphone, hydrocodone, oxymorphone, and oxycodone derivatives of morphine and codeine could be selectively separated from their associated codeine and morphine parents. 6-MAM, the primary metabolite of heroin, was also included.

HILIC parameters including buffer, pH, and concentration were evaluated on both bare silica and zwitterion-HILIC columns. Acetone and acetonitrile organic modifiers were evaluated as well. Analyte separation was achieved using the zwitterionic stationary phase with a mobile phase composed of 85% acetone and 15% ammonium formate buffer (5mM with pH of 5).

Solid-Phase Extraction (SPE) with a C18 sorbent was used to improve signal-to-noise ratio and recovery. This required a two-step elution involving a more aprotic solvent to elute the glucuronides. Electrospray Ionization (ESI) Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS) was used in positive Multiple Reaction Monitoring (MRM) mode. Transitions used to evaluate M6G and M3G were 462/286 and 462/201.

It was discovered that retention and selectivity were highly influenced by how nonpolar and aprotic the organic modifier was in the mobile phase. The HILIC mechanism relies on the partition created between the stagnant water layer and the organic mobile phase. By reducing the polarity of the organic modifier, a more defined partition was created and allowed for different

selectivity. The bare silica stationary phase did not provide the selectivity needed for both structural isomers and hydromorphone/hydrocodone derivatives simultaneously. Because M3G/M6G, hydrocodone/codeine, and hydromorphone/morphine pairs share the same molecular ion, retention data is needed to qualitatively identify each analyte. Retention time data Coefficient of Variations (CVs) as low as 2% demonstrated reproducibility for the qualitative determination of each analyte. Limits Of Detection (LOD) as low as 5ng/mL and Limits of Quantitation (LOQ) of 10ng/mL were observed for the morphine glucuronides. Hydromorphone and hydrocodone provided good signal-to-noise with LOD values as low as 2ng/mL and LOQ values of 10ng/mL.

The mostly organic mobile phase proved highly compatible with ionization in the ESI interface of the triple quadrupole detector. The sample preparation and compatibility to the detector allowed for a working linear range of 1ng/mL – 2,000 ng/mL with R² values close to 1. This method provides separation of hydrocodone and hydromorphone from parent opiates which is often overlooked when evaluating selectivity of a new morphine and codeine method.

HILIC, Opiates, Glucuronides

K11 A Two-Month Stability and Distribution Study of the Benzodiazepine Phenazepam

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After attending this presentation, attendees will better understand the extent to which 7-bromo-5-(2-chlorophenyl)-1,3-dihydro-2H-1,4-benzodiazepin-2-one (phenazepam) undergoes postmortem redistribution and its stability in blood and urine.

This presentation will impact the forensic science community by raising awareness concerning phenazepam distribution, stability, and the subsequent interpretation of toxicological results. As this drug is emerging as a controlled drug in Europe and a drug of concern in the United States, laboratories need to be aware of the concentration among biological specimens that can be encountered. Also, the stability study will aid in accounting for postmortem redistribution and putrefaction of the drug.

Outside of the United States, phenazepam has been prescribed as an anxiolytic, anticonvulsant, and sedative since its development in Russia in the 1970s. Compared to other 1,4-benzodiazepines, it is more potent and longer lasting with a half-life up to 60 hours. It has been recently controlled in parts of Europe and, although it has been declared a drug of concern in the United States, only a very small number of states have controlled its use.¹ Pharmacological studies on phenazepam are extremely limited and little is understood regarding its distribution in postmortem samples. In this study, the postmortem distribution of phenazepam is reported in a series of ten toxicology cases. Stability issues will also be addressed in blood and urine.

The distribution of phenazepam in postmortem femoral, subclavian and central blood, antemortem blood, urine, vitreous humor, and skeletal muscle was determined using liquid-liquid extraction and liquid chromatography-tandem mass spectrometry. The concentration ranges were as follows: 0.010-0.272mg/L for femoral blood; 0.024-0.171mg/L for subclavian blood; 0.202mg/L for antemortem blood; 0.067mg/L for chest blood; 0.001-0.030mg/L for urine; 0.001-0.016mg/L for vitreous humor; and 0.023-0.522mg/kg for skeletal muscle. Highest concentrations of phenazepam

were observed in the skeletal muscle and the lowest concentrations were in the vitreous humor. Highest blood concentrations were seen in antemortem blood which were 6-10 times greater than urine and vitreous humor.

For the stability study, drug-free whole blood and urine were fortified with phenazepam (0.25mg/L) and stored at room temperature (20°C) and refrigerated (4°C) for two months. Samples were extracted at day 0, 1, 3, 7, and then weekly for the remainder of the two-month study. There was very little change in concentration over two months. Days 7 and 35 marked significant points in the concentration trend. On day 7, there was an overall decrease in concentration, and after day 35, there was an increase in concentration over all the fluids except blood at room temperature. There was a noticeable concentration variation between the specimens after day 35. However, at day 56, all the fluids had a concentration within 10% of each other. From the original day 0 concentration, the decrease in concentration ranged from 16-30%.

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Phenazepam, Stability, Distribution

K12 Identification of *In Vitro* Metabolites of PB-22 and 5-F-PB-22 by UPLC-QTOF Mass Spectrometry

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After attending this presentation, attendees will be able to identify major *in vitro* metabolites of the novel cannabimimetic compounds PB-22 and 5-F-PB-22. The goal of this presentation is to present an Ultra Performance Liquid Chromatography-Quadrupole Time-Of-Flight/Mass Spectrometry (UPLC-QTOF/MS) method for the identification of *in vitro* metabolic transformations.

This presentation will impact the forensic science community by presenting a qualitative analytical method for the detection of PB-22 and 5-F-PB-22 metabolites and identification of suitable markers for use.

Cannabimimetic compounds are sprayed onto plant material and sold for use as recreational drugs despite their adverse effects. Two compounds that have been identified in herbal products are quinolinyl carboxylates, PB-22 and 5-fluoro PB-22.

In vitro samples were generated by incubating the compounds at 100µM in cryopreserved human hepatocytes. At each of three distinct time points, 0, 15, and 120min, an aliquot of 100µL was removed and quenched with acetonitrile containing 0.2% acetic acid. A portion of each aliquot was hydrolyzed with 25µL of 12.3 units/µL of beta-glucuronidase in 0.1M ammonium acetate and incubated for 3.5h at 60°C.

Data were collected on both the hydrolyzed and unhydrolyzed samples using a Waters® Acquity UPLC® interfaced to a Waters® Synapt® G2 QTOF mass spectrometer. All data were acquired using a Mean Squared Error (MS^E) method, which acquires both low- and high-energy data simultaneously. Liquid chromatography was carried out using an Acquity BEH C18 column (1.7µm x 2.1 x 50mm) connected to a Vanguard BEH C18 pre-column (1.7µm x 2.1 x 5mm) and held at 30°C. A gradient elution with a flow rate of 500µL/min was used with mobile phase

A consisting of water with 0.1% formic acid and mobile phase B consisting of acetonitrile with 0.1% formic acid.

In positive ion electrospray mode, PB-22 and 5-F-PB-22 parent compounds and metabolites were present as both protonated and sodiated ions. Major metabolic transformations observed for both compounds were 3-carboxyindoles and their corresponding glucuronide conjugates. Hydroxylated PB-22 and defluorinated 5-F-PB-22 followed by hydroxylation were also identified. Interestingly, no evidence for hydroxylated 5-F-PB-22 was observed. These results are in contrast to the metabolism of other currently prevalent cannabimimetic compounds in which the dominant metabolites result from monohydroxylations.

This research provides forensic practitioners with the identification of major *in vitro* metabolites of novel designer drugs PB-22 and 5-F-PB-22. Major metabolic transformations are similar for both compounds. Due to defluorination of 5-F-PB-22, several metabolites are common to both compounds, and therefore should not be used for unambiguous identification.

PB-22, 5-F-PB-22, Metabolites

K13 Determination of GHB in Blood and Urine Using GC/MS/MS Without Derivatization

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After attending this presentation, attendees will learn about a new method for determining concentrations of Gamma-Hydroxybutyric Acid (GHB) and Gamma-Butyrolactone (GBL) in toxicology specimens by Gas Chromatography Tandem Mass Spectrometry (GC/MS/MS), without the need for derivatization. Compared to ethanol, GHB is a more powerful central nervous system depressant that can be used and abused for multiple purposes with deadly consequences. Forensic toxicology laboratories that analyze specimens from Driving While Intoxicated (DWI) suspects, victims of Drug-Facilitated Sexual Assault (DFSA), and postmortem investigations need a durable method for GHB/GBL analysis to invoke when necessary.

This presentation impacts the forensic science community by relaying a novel procedure that utilizes the sensitivity and selectivity of GC/MS/MS, while promoting a more routine workflow by negating the need for hazardous derivatization agents. GHB is a controlled substance used for medicinal therapy of cataplexy, insomnia, and fibromyalgia. Because of its intoxicating effects, GHB can be abused in bodybuilding, lowering inhibitions or rendering a victim defenseless in DFSA, and as a recreational club drug. GHB is a relatively small polar molecule that can be difficult to detect and is rapidly eliminated from the body after ingestion. Low levels of endogenous GHB are common in all mammals, complicating detection and toxicological interpretation.

Derivatization is a common method for GHB analysis; however, derivatizing agents increase costs, are harmful to gas chromatography columns, and are hazardous to the analytical chemists who use them. Acid-catalyzed conversion of GHB to GBL is a much cleaner and less expensive way to analyze GHB. In the presence of concentrated sulfuric acid, GHB converts from its linear polar form to GBL, a less polar-cyclic lactone which is less damaging to the GC column since it does not need to be derivatized. By avoiding derivatization, both time and money are saved by cutting out extra steps with derivatizing agents. In this method, extracts of GHB from the sample are split into cyclized and non-cyclized preparations for analysis. Any GBL already present in the sample will be revealed by its inherent volatility, while the

remaining non-cyclic GHB is converted to GBL using sulfuric acid and therefore detected as GBL in the acquisition method.

Specifically, a liquid-liquid extraction employing methylene chloride was used to extract blood and urine samples fortified with varying concentrations of GHB. This novel method produced an average R^2 value of 0.99 in urine ($n=3$) and 0.99 in blood ($n=3$). The range of the calibration curve was linear from 10mg/L to 400mg/L of GHB in blood and urine. The acquisition method involved a splitless injection of 1 μ L of extract at 250°C into a DB35 ultra inert GC column. The oven program was set to an initial temperature of 50°C for 2 minutes, then increased at a rate of 50°C/min to 280°C with a hold of 6.5 minutes. The flow rate was set to 1.5mL/min with a total run time of 13.1 minutes. A modest 10V collision energy potential was sufficient to produce optimal fragmentation ions. The transition product ions for GBL were identified at 86m/z to 42m/z for quantitation and 86m/z to 39m/z for qualifier ions. Due to its similar size and properties, delta-valerolactone was used as an internal standard for this analysis. By using this non-deuterated internal standard, the R^2 value of the calibration curve only needed to be above .98 to be acceptable. The transition product ions of delta-valerolactone were determined to be 100m/z to 41.1m/z for quantitation and 100m/z to 56.1m/z for qualifier ions.

The method was validated in blood and urine for routine screening of DFSA casework, as well as evaluating DWI suspects and postmortem cases. Other published studies have recommended a 10mg/L cutoff to accommodate a low level of GHB while recognizing a level above endogenous concentration. Meanwhile, the upper limit of quantitation extends well into the range of blood concentrations reported to cause fatal toxicity. This procedure will reduce costs and improve time management, while providing a sensitive and selective method for GHB analysis.

Gamma-Hydroxybutyric Acid, Gas Chromatography Tandem Mass, Gamma-Butyrolactone

K14 Development of Sensitive Polyclonal Antibodies for the Detection of the Adamantanylacetylindole Family (AB-001) and the Phenylacetylindole Family (RCS-8 and JWH-250) of Synthetic Cannabinoids

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After attending this presentation, attendees will understand the development of the first highly sensitive and generic polyclonal antibodies for the detection of the adamantanylacetylindole family (AB-001) and the phenylacetylindole family (RCS-8 and JWH-250) of synthetic cannabinoids for application to the development of immunoassays that are not currently available. The goal of this presentation is to expand the screening of synthetic cannabinoids in drug testing.

This presentation will impact the forensic science community by demonstrating the development of the first polyclonal antibodies for the detection of AB-001, RCS-8, and JWH-250 of

synthetic cannabinoids for the development of new immunoassays applicable to drug testing.

Background: Since their introduction into the marketplace in 2004 as "legal highs," synthetic cannabinoids have rapidly increased in popularity in the United States and Europe. Originally sold under the brand name "Spice," this brand name has become a generic term to include the entire class of "legal" smoking blends sold on the internet. Such compounds act as CB1 cannabinoid receptor agonists and have a psychophysiological action similar to tetrahydrocannabinol (THC). Following the identification of the first members of this drug class in 2008, a number of countries introduced legislative controls to make them illegal. More than 50 synthetic cannabinoids have been detected and identified in legal high products to date. It is expected that many more will follow in an attempt to overcome legislation bans. One recently identified cannabimimetic is 3-((adamantan-1-yl) carbonyl)-1-pentylindole (AB-001), which is structurally similar to AM-1248 (1-((N-methylpiperidin-2-yl)methyl)3-adamant-1-yl)indole and 5-fluoropentyl 5-F-AB-001. Urinary metabolites of AB-001 have been recently identified and the parent compound was reported to be absent in urine.¹ 1-(2-cyclohexylethyl)-3-(2-methoxyphenylacetyl) (RCS-8) indole is a synthetic cannabinoid (also known as SR-18 or BTM-8). It can be described as an analogue of JWH-250 with the 1-pentyl group replaced by 1-(2-cyclohexylethyl) and has been recently reported as less potent than JWH-018, SDB-001, and Δ^9 -THC.² Despite not having been reported in the scientific or patent literature as yet, reputed recreational use of RCS-8 in the United States has led to it being specifically listed in a proposed 2011 amendment to the Controlled Substances Act, which is proposing to add a number of synthetic drugs into Schedule I.

Methods: The immunogens comprising AB-001 hapten conjugated to bovine thyroglobulin (BTG) and RCS-8 hapten conjugated to BTG as a carrier protein were administered to adult sheep on a monthly basis to provide target-specific polyclonal antiserum. Immunoglobulin G (IgG) was extracted from the antiserum and evaluated via competitive immunoassay, specifically Enzyme-Linked Immunosorbent Assay (ELISA).

Results: Polyclonal antibody against AB-001: in this initial evaluation, the assay was standardized to the metabolite AB-001 N-pentanoic acid. The developed polyclonal antibody also detected the metabolite AB-001 N-pentanol (cross-reactivity: 40%) and presented a cross-reactivity of 7% with the parent compound. The antibody did not cross-react with the first generation of cannabinoids JWH-018, JWH-073, and JWH-250 (cross-reactivity: <3.7%) and presented cross-reactivity >10% with UR-144 N-pentanoic acid, UR-144 N-pentanol, and XLR-11. The sensitivity, expressed as half maximal inhibitory concentration (IC_{50}), was 0.7ng/ml. Polyclonal antibody against phenylacetylindole family (RCS-8 and JWH-250): in this initial antibody evaluation, the assay was standardized to JWH-250, the antibody also showed >25% cross-reactivity with RCS-8. The IC_{50} value was <0.5ng/ml for a calibration range 0-50ng/ml.

Conclusion: This initial evaluation shows the development of the first highly sensitive polyclonal antibodies for the detection of adamantanylacetylindole and phenylacetylindole synthetic cannabinoids. The results indicate that these antibodies are suitable for the development of immunoassays for the detection and quantification of these compounds in human samples with important forensic and toxicological applications.

References:

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Apetz N., English K., Brooker L., Goebel C., Hibbs D.E., Glass M., Connor M., McGregor I.S., Kassiou M. The synthesis and pharmacological evaluation of adamantane-derived indoles: cannabimimetic drugs of abuse. *ACS Chem. Neurosci.* 2013

Synthetic Cannabinoids, Generic Antibodies, Immunoassays

K15 Determination of Lead and Zinc in Urine by Graphite Furnace and Flame Atomic Absorption Spectrophotometry (GFAAS) in Saudi Populations

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After attending this presentation, attendees will better understand the use of Graphite Furnace and Flame Atomic Absorption Spectrophotometry (GFAAS) for metal urinalysis.

This presentation will impact the forensic science community by comparing urinary lead and zinc concentrations in different populations.

Heavy metals enter biological fluids through many sources including food, smoking (tobacco smoke), and by intake of contaminated liquids. In recent years, tobacco smoking has become one of the major sources of intake of heavy metals. It has been shown that Environmental Tobacco Smoke (ETS) is a complex mixture of chemicals found in air as a specific result of smoking (side stream smoke that is released from the lit end of the cigarette and exhaled main stream smoke that is exhaled by the smoker after drawing on the cigarette). This study compares the concentration of lead and zinc taken from smokers, passive smokers, and non-smokers. It also makes comparisons with the international maximum allowed limits, and the correlation between concentrations of these elements and the duration of smoking.

Heavy metals like lead and zinc (Pb and Zn) were determined in urine samples obtained from 67 volunteers (smoker, non-smokers, and passive smokers) from different districts of the city of Riyadh, Saudi Arabia. Urine samples were filtered from 0.45 μ MS[®] Nylon Filter, then 10 microliters of concentrated nitric acid (HNO₃) per 1ml of urine sample were added. After collecting the samples, they were stored in the refrigerator at -20°C prior to analysis. Five milliliters of the urine sample were digested with nitric and hydrochloric acid. Analysis was performed using GFAAS for lead and Flame Atomic Absorption Spectrophotometry (FAAS) for zinc. Accuracy of the analysis was confirmed by analyzing control samples. The results proved that the concentration of lead and zinc were high in smokers and passive smokers. The concentration of lead in smokers was between 91.03-185.54 μ g/l, which is higher than the international maximum allowed limits (0.01-2.14 μ g/l). The concentration of zinc in smokers was between 0.42-1.03mg/l, which is higher than the international maximum allowed limits (0.44-0.499mg/l). Therefore, smoking may be the reason for increased concentrations of heavy metals in smokers' urine. This study revealed that the level of lead and zinc was related to the smoking duration. The concentration of heavy metals (lead and zinc) was higher in smokers' and passive smokers' urine compared to non-smokers, and a correlation was found between concentration and length of time spent smoking.

GFAAS, Lead, Urine

K16 Hair Analysis as a Tool to Evaluate the Prevalence of Synthetic Cannabinoids in Different Populations of Drug Consumers

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After attending this presentation, attendees will learn how to investigate the current diffusion of synthetic cannabinoids among the population by means of hair analysis.

This presentation will impact the forensic community by describing which molecules among new synthetic cannabinoids are the most consumed in the Piedmont Region of Northern Italy, and by profiling the typical consumers of these new psychoactive substances.

Forensic laboratories are challenged worldwide with the detection of New Psychoactive Substances (NPSs) in different biological specimens, including hair. Among the new psychoactive products, herbal mixtures containing synthetic cannabinimetics are likely the most abused worldwide. Although the consumption of these drugs has been tolerated, today the majority of them are progressively banned. Still, serious side-effects and acute intoxications are becoming increasingly frequent.

Several analytical methods were developed to determine some cannabinimetics and/or their metabolites in either blood or urine, but no routine urine screening test is generally performed in forensic laboratories globally to detect them on large population sets. Consequently, almost no prevalence studies are available. For the same reason, the potential association between NPS use and traffic or occupational accidents is unknown.

In this study, a specific Ultrahigh Pressure Liquid Chromatography/Tandem Mass Spectrometry (UHPLC/MS/MS) method for the detection of 23 synthetic cannabinoids in hair samples was developed in order to: (1) expand the number of screened compounds, including new substances emerging in the European territory; (2) evaluate their consumption over a long period of examination (year 2011); and, (3) evaluate the prevalence of cannabinimetics among different populations of drug consumers. The method employs alkaline digestion of hair samples followed by extraction with n-hexane/ethyl acetate, and injection into the UHPLC/MS/MS system. After validation, the method was applied to the analysis of 344 hair samples tested in 2011 in the laboratory for the most common drugs. Overall, 15 samples (4.4%) were found positive for at least one synthetic cannabinoid. Consistent with previously published results, the present data show that young males, who are former or still active cannabis consumers, represent the population most often involved in synthetic cannabinimetics consumption. Several cases of poly-abuse were also determined. The drug most frequently detected was JWH-073 (11 samples), generally at low concentration (mean 7.69 ± 14.4 pg/mg, median 1.9 pg/mg, range 1.6-50.5 pg/mg), followed by JWH-122 (8 samples, mean concentration: 542 ± 962 pg/mg, median 28.4 pg/mg, range 7.4-2784 pg/mg). Other detected drugs included JWH-250, JWH-081, JWH-018, JWH-210, JWH-019, and AM-1220. For several positive samples, the synthetic cannabinoid concentration was lower than 50 pg/mg, supporting the need of established cut-off values for discrimination between chronic consumption and occasional use (or external contamination).

Similarly to what has happened in the last two decades for tetrahydrocannabinol (THC), further studies of synthetic cannabinoids are needed to establish: (1) the presence of metabolites to prove consumption; (2) the concentration ratios

between parent drugs and metabolites; and, (3) the identification of alternative markers in order to support the interpretation of hair analyses.

Hair, Synthetic Cannabinoids, Spice

K17 Poisoning Trends in Nepal

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After attending this presentation, attendees will gain an understanding about poisoning trends in developing countries, as well as understanding which poisons are used in the majority of poisoning cases.

This presentation will impact the forensic science community by providing results from a ten-year retrospective study in Nepal with very little previous research. This presentation will add to research being carried out in forensic toxicology by broadening the understanding of controlling the spread of toxic substances and by analyzing for better and accurate results of these poisons.

In 16 high-income and middle-income countries, poisoning is the fourth largest cause of unintentional injury, after traffic injuries, fires, and drowning. Fatal poisoning rates in low-income and middle-income countries are four times that of high-income countries.

Common poisoning agents in high-income countries include pharmaceuticals, household products, pesticides, poisonous plants, and bites from insects and animals. This presentation is a ten-year retrospective study evaluating poisoning cases in Nepal on the basis of data from the National Forensic Science Laboratory and Central Police Forensic Science Laboratory, Nepal. The laboratories received 11,596 cases (36,384 samples) for toxicological analysis during the years 2000 to 2012. Out of 11,596 cases, 3,029 cases were found positive for poisoning.

Statistics from the last decade show that out of 3,029 poisoning cases, 44% of the cases were the result of consuming insecticides and 11% of the cases were rodenticides. The remaining 45% of the cases were due to drugs, plant poison, alcohol, acid, carbon monoxide, etc. In these cases, female victims outnumbered male victims.

During the last fiscal year (2012-13), the laboratory received 1,668 cases of suspected poisoning. Most of the suspected cases of poisoning are the result of suicide and homicide. Accidental poisoning is rare. The trends of poisoning show that there are more cases in urban areas compared to rural areas. However, the poisoning cases are on the rise from year to year. Fatal and non-fatal poisonings are strongly associated with lower socioeconomic status, between and within countries. The prevalence and types of poisoning vary in different parts of the world, depending on industrial development, agricultural activities, cultural practices, and customs. Other risk factors for poisoning include those related to the poisoning agent itself, including: toxicity; nature; physical appearance and storage; season and weather conditions; policies, standards and laws governing the manufacture, labeling, distribution, storage, and disposal of poisoning agents; and access to quality health care for treatment.

This study showed that poisoning trends have increased since 1988 when the first poisoning case was investigated by the Toxicological Division of National Forensic Science Laboratory, Nepal, indicating a grave problem that needs to be solved.

Poisoning, Retrospective Study, Nepal

K18 LGC Quartz Forensic Blood Toxicology Proficiency Testing Scheme: A Review

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After attending this presentation, attendees will appreciate the value and utility of proficiency tests within forensic toxicology. Attendees will also be encouraged to perform in-depth review of their proficiency test results.

This presentation will impact the forensic science community by improving laboratory operation and providing laboratories with greater awareness of what constitutes an appropriate test protocol and the significance of performance scores.

Proficiency testing should guard against laboratories having undetected errors and may additionally provide educational benefits. LGC Standards® is an international, United Kingdom Accreditation Service- (UKAS) accredited provider of commercial Proficiency Testing Schemes (PTS). Their scheme relevant to forensic toxicology is the Forensic Blood Toxicology (QUARTZ) PTS, in operation for more than ten years. This presentation provides a long-term review of QUARTZ to evaluate the effectiveness of the scheme for detecting analytical bias and for improving laboratory performance, its overall fitness-for-purpose. Review of a fitness-for-purpose scheme should establish an indication of the level at which participants are performing.

Method: Results of the PTS from round 30 in 2007 to round 48 in 2012 were provided by the coordinators in the form of the summary reports provided to participants. In each round, there were both qualitative and quantitative assessments. The results from both were collated in spreadsheets for data grouping and trend analysis. Quantifications did not have a consistent scoring system across all rounds. Scoring methods were investigated and all participant results were awarded a z-score using a provided assigned value and a percentage of this value as acceptable deviation.

Results: Nineteen rounds of QUARTZ spanning six years were summarized. There was a regular group of participants, with minor fluctuations, averaging 18 per round, range 16-21. Gas Chromatography-Mass Spectrometry (GC/MS) was the most-used analytical method. Detailed trend analysis was not possible with the QUARTZ scheme due to the extensive analyte menu and wide variety of analytes tested. There was some repetition of morphine analysis which indicated difficulties requiring further investigation. It was not possible to determine the level at which participants generally perform.

Conclusion: QUARTZ is a good educational resource. Laboratories that establish proficiency in their validated methods elsewhere can benefit from the varied nature of this scheme to gain experience with analytes which are not encountered often or which are emerging substances of abuse. However, in order to provide a safeguard against bias, this scheme requires more repetitive testing, which would assist in providing an effective means of monitoring routine laboratory performance. It is recommended that a selection of the most commonly encountered analytes in forensic toxicology (e.g., morphine, codeine, diazepam, cannabis, amphetamine, cocaine, and methadone) be included at least once per annum.

Proficiency Scoring, Proficiency Testing, Forensic Toxicology

K19 An Internet-Directed Hydrogen Sulfide Suicide: Fast Fatality in Northeast Ohio

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After attending this presentation, attendees will better understand poisoning by hydrogen sulfide as a suicide method.

This presentation will impact the forensic science community by informing forensic professionals about: (1) a novel suicide technique found on the internet used particularly by our youth; (2) concerns for first responders; (3) unique pathology findings; and, (4) providing a discussion of toxicology results of this substance.

Suicide from internet-directed hydrogen sulfide (H₂S) asphyxiation has been seen in the United States since August 2008, following a trend observed in Japan earlier that year. Instructions posted on the internet include which cleaners and pesticides to buy, how to mix these commercially available chemicals to create the gas, and on some sites, a note to print out that cautions first responders to the presence of H₂S gas.

Hydrogen sulfide is poisonous primarily in the gaseous form, which is characterized by a "rotten egg smell" at low concentrations. Continued exposure to H₂S results in desensitization of the olfactory senses so that it is no longer detected. Normal healthy subjects have thiosulfate levels of less than 0.3mg/L. Sulfide in the body is partially converted to sulfate and thiosulfate through oxidization. Baselt recommends that thiosulfate levels should be assessed to establish fatal intoxications. H₂S inhibits cellular respiration by binding with the iron of cytochrome oxidase, in the same fashion as hydrogen cyanide, thereby inhibiting binding of oxygen and stopping cellular respiration, resulting in death.

A 26-year-old White male was found in his car with a note taped to the window reading "Stay away! poisonous [sic] chemicals! call [sic] 911". Also in the car were a bucket, bottles of toilet bowl cleaner and lime sulfur spray, and two other notes, both indicating that H₂S gas had been formed. A reading of 8 parts per million H₂S gas was taken by responding authorities. HAZMAT removed a 5-gallon bucket with liquid that was located in the front passenger seat and quickly made it neutral. Suicide notes were found addressed to his family.

A complete forensic autopsy was performed at the Cuyahoga County Medical Examiner's Office (CCMEO), Cleveland, Ohio. It consisted of an external and internal examination of the body, microscopic examination, and toxicological examination. Samples of heart and femoral blood, vitreous humor, gastric contents, bile, urine, and liver were submitted for comprehensive toxicological examination.

The body was that of a normally developed 26-year-old adult White man with a length of 64 inches (5'4") and a weight of 157 pounds. External examination was significant for distinct gray/green discoloration of the face, lips, conjunctivae, abdomen, hands, and feet. Internal examination revealed mild, generalized brain edema with diffuse green discoloration of the cortex and deep gray structures. There was marked dark green discoloration of the substantia nigra, red nuclei, and cerebellar dentate nucleus. The cerebral and cerebellar white matter was unremarkable. Visceral organs appeared normal grossly. Microscopic examination revealed diffuse brain and visceral congestion with focal perivascular acute

necrosis in the brain, evidence of acute microscopic cardiac ischemia, and hemorrhagic pulmonary edema.

Toxicological testing revealed ethanol 0.02g/dL, caffeine, and cotinine in the femoral blood. Heart blood was sent to AIT Laboratories in Indianapolis, Indiana, for a comprehensive drug screen and to National Medical Services (NMS) Labs in Willow Grove, Pennsylvania, for thiosulfate analysis. AIT Laboratories found the femoral blood positive for alprazolam 14.3ng/mL and caffeine. NMS Labs reported 43mg/L thiosulfate in the decedent's femoral blood, which is 143 times higher than the thiosulfate level of a healthy subject.

The autopsy findings in this case were consistent with the suspicion that death was due to acute exposure to H₂S. The cause of death was determined to be asphyxia due to hydrogen sulfide inhalation. The manner of death was ruled as suicide.

Hydrogen Sulfide, Suicide, Internet

K20 Qualitative Screening of Multiple Designer Drug Classes Using Polymer-Based SPE and LC/QTOF/MS

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After attending this presentation, attendees will understand the advantages and limitations of using high-resolution Tandem Mass Spectrometry (MS/MS) for screening of designer drugs from multiple classes. Attendees will also learn about the advantages of using polymeric solid phase extraction cartridges for screening of designer drugs. In addition, the presentation will demonstrate the applicability of a high-resolution MS/MS screening method in combination with a previously created high-resolution MS/MS spectral library.

This presentation will impact the forensic science community by providing a high-resolution MS/MS method using a polymer-based solid phase extraction cartridge to qualitatively screen for more than 200 designer drugs from classes including cathinones, indanes, phenethylamines, piperazines, tryptamines, and synthetic cannabinoids.

Designer drugs are compounds that are used in an attempt to evade current drug laws. Many of these compounds are structurally or pharmacologically similar to common illegal drugs of abuse. In the past few years, there has been an increase of recreational designer drug use among users. In an effort to combat designer drug use, the U.S. government permanently scheduled 26 designer drugs in the Synthetic Drug Abuse Prevention Act of 2012. Due to potential dangers and legal implications that arise from the use of such drugs, it is critically important that forensic toxicology laboratories have the capability to screen and detect as many designer drugs as possible in a single specimen.

In order to address the dynamic nature of designer drug use and to screen for these drugs effectively, many laboratories have turned to mass spectrometric screening techniques. Liquid Chromatography/Quadrupole Time-Of-Flight/Mass Spectrometry (LC/QTOF/MS) was used for this project because it enables the analyst to have higher confidence when identifying a compound, due to its high resolution, high mass accuracy, and MS/MS capabilities. The LC/QTOF/MS also has high sensitivity in full-scan mode, which is useful for screening designer drugs. This project focused on creating and validating a screening method

that employs a Bond Elut® Plexa™ PCX Solid-Phase Extraction (SPE) cartridge, which is a polymer-based cation mixed-mode cartridge and an Agilent® 6530 Accurate-Mass QTOF LC/MS. The solid phase extraction method for this project was created after optimizing the load, wash, and elution steps in order to achieve the highest percent recovery of the designer drugs. Different volumes of sample and wash/elution steps were investigated to determine which volumes would produce the lowest Limit Of Detection (LOD) while minimizing the interferences that may be present in the post-extraction solution. The LC gradient, auto MS/MS parameters, and library search parameters were optimized to produce a method that generated the lowest amount of false positives while also avoiding false negatives. Once the method was optimized, it was then validated using Scientific Working Group for Forensic Toxicology (SWGTOX) recommendations. The LOD for most of the designer drugs was <10ng/mL. No significant carryover or interference was observed. Ionization suppression and enhancement fell in the acceptable range (±25%).

The results demonstrated that the use of a polymer-based SPE cartridge and high-resolution LC/QTOF/MS instrumentation can be useful tools in comprehensive screening for designer drugs. The larger binding capacity of the polymer-based SPE cartridge combined with the high resolution of the LC/QTOF/MS enables higher confidence in drug identification when screening for designer drugs. While there are limitations to this screening method, i.e., some isobaric and poorly ignitable compounds in Electrospray Ionization (ESI) mode, the method represents a new, useful tool for the detection of designer drugs in human matrices.

Designer Drugs, LC/QTOF/MS, SPE

K21 Evaluation of Postmortem Methamphetamine Concentrations: A Series of Case Studies

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The goal of this presentation is to report and describe observed postmortem blood methamphetamine/amphetamine concentrations in a series of cases within the context of case history, Cause Of Death (COD), and Manner Of Death (MOD) determinations. The expected outcome is a comparison and contrasting of blood methamphetamine/amphetamine concentrations between cases in which the drug and metabolite were deemed to be COD or contributing-cause related and cases in which the methamphetamine/amphetamine were considered incidental to COD.

This presentation will impact the forensic science community by augmenting previously reported blood concentrations of methamphetamine/amphetamine in forensic toxicology references described for postmortem cases.

Incidents involving illicit methamphetamine, also known as "meth," "crystal," "ice," "glass," and "speed" have demonstrated a dramatic increase in recent years. This prevalence has been attributed to the low cost, availability, and extended duration of action as a central nervous system stimulant. Furthermore, the ease of production from inexpensive and readily available materials has contributed to its increased production and distribution throughout the United States. When used moderately, effects include euphoria, increased heart rate, elevated blood pressure, reduced appetite, and loss of inhibition. Excessive or chronic use may lead to manic and violent behavior, dizziness, confusion, hyperthermia,

seizure, cardiorespiratory depression, and death. Typical routes of administration include insufflation, smoking, ingestion, and intravenous injection.

The pharmacological properties of methamphetamine are well documented. The drug works as a dopaminergic central nervous system stimulant by freeing dopamine from nerve terminal storage vessels and blocking its reuptake while also endorsing the release of norepinephrine and inhibiting its reuptake. The physiological effects of long-term use include tooth decay, parasitosis, fatigue, and malnutrition. Several studies have also indicated a strong association between methamphetamine use and cardiac complications (coronary artery disease, hypertension, cardiac enlargement, etc.) as well as subarachnoid hemorrhage. In addition, numerous studies have been reported on postmortem methamphetamine concentrations.

Despite this, debate remains in regard to cause-of-death determinations based on methamphetamine presence and postmortem toxicology results. Presented here are a series of cases submitted to this study's laboratory for toxicology testing. Analyses for methamphetamine were completed on postmortem blood specimens using Liquid Chromatography with Time-Of-Flight Mass Spectrometry (LC/TOF) followed by confirmation using Liquid Chromatography with Tandem Mass Spectrometry (LC/MS/MS). Cases were selected from a pool of specimens that resulted in the detection of methamphetamine submitted to the laboratory over a 12-month period (January 1, 2012-December 31, 2012).

Examination of the methamphetamine concentrations determined in each case, along with case histories for each decedent, demonstrate considerable overlap in methamphetamine concentrations related to deaths from intoxication and those related to other causes and manners (suicide, homicide, traffic accidents, cardiovascular incidents, etc.). For cases where the cause of death was due solely to methamphetamine intoxication, postmortem blood concentrations ranged from 102ng/mL to 17,662ng/mL. Considerable variation was also seen for those cases in which methamphetamine was determined to be an incidental finding, with a range from 91.5ng/mL to 9.10 ng/mL seen. For each specimen, this presentation discusses the case history, toxicology findings, and autopsy findings (if performed) with the objective of demonstrating the diversity of postmortem methamphetamine concentrations relative to the actual cause-of-death determinations.

Toxicology, Postmortem, Methamphetamine

K22 Resources for Responding to the Challenge of Emerging Drugs: Where Is the Information Your Analysts Need?

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After attending this presentation, attendees will understand the forensic resources available to help assist analysts in identifying spectral data of emerging drugs.

This presentation will impact the forensic science community by providing an overview of the growing array of

information and resources available to aid in the identification and interpretation of data related to emerging drugs.

Forensic laboratories are faced with a challenge when attempting to identify unknown compounds in a sample. The rise in prevalence of new designer drugs such as "Spice" and "bath salts" has increased this challenge as uncontrolled drug analogs rapidly appear on the drug market. There exists a need in the community to provide laboratory personnel with assistance in identifying these emerging designer drugs as many of these compounds have yet to be identified and characterized in the literature.

In October 2012, the National Institute of Justice (NIJ) sponsored an analog working group meeting at which professionals representing several disciplines of forensic science were present. Attendees included commercial vendors, research institutes, federal, state, and local crime laboratories, DEA, and customs. The group discussed major challenges that forensic laboratories are faced with when trying to identify emerging drugs. Input on the most valuable resources utilized by these attendees was recorded and compiled for distribution in addition to resources identified by Response To Intervention (RTI) scientists. Resources include downloadable Electron Ionization/Mass Spectrometry (EI/MS) spectral libraries such as the Scientific Working Group for the Analysis of Seized Drugs (SWGDRUG), the American Academy of Forensic Sciences (AAFS), and Cayman Chemical mass spectral library, all of which are freely available to download online. Other downloadable resources include commercial spectral libraries that include tandem MS data and EI/MS spectral data. There are free, Web-accessible and searchable databases, such as ForensicDB, that include multiple spectral methods, structures, and instrumental parameters. Other resources include drug monographs, peer-reviewed spectral data, and active discussion forums. All of these resources provide the forensic community with spectral data for comparison and other helpful information to assist with unknown compound identification.

Although several resources are available, the challenge remains to adequately disseminate this information to the forensics community. Consolidation of these resources would allow valuable information to become available in multiple formats, therefore reaching a wider audience.

Forensic Resources, Spectral Databases, Designer Drugs

K23 Analysis and Detection of Amphetamine-Type Stimulants and Ketamine in Rat Hair Using SPE/GC/MS

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After attending this presentation, attendees will understand the potential for the simultaneous extraction of amphetamine, methamphetamine, mephedrone, methylone, and ketamine from rat hair samples using Solid Phase Extraction (SPE). In addition, attendees will be aware of how to detect these compounds using Gas Chromatography/Mass Spectrometry (GC/MS).

This presentation will impact the forensic science community by demonstrating a method for the simultaneous extraction and detection of amphetamine-type stimulants and ketamine in rat hair, which can also be applied to human hair. As these drugs are currently banned, it is important that laboratories develop assays that can simultaneously extract and detect several compounds in order to reduce laboratory expenses of performing separate assays.

Marketed as “legal highs,” the amphetamine-type stimulants, such as mephedrone and methylone, are central nervous system stimulants which produce similar effects to amphetamine. Ketamine, which was marketed as an anesthetic, became a recreational drug causing similar effects to phencyclidine (PCP). The commonality between the amphetamine-type stimulants and ketamine is the high-abuse potential associated with taking them. The high-abuse potential among these compounds causes the need for laboratories to have a quick, single assay for the extraction and detection of these compounds among users.

The objective of this research was to apply a SPE method to simultaneously extract amphetamine, methamphetamine, mephedrone, methylone, and ketamine from rat hair samples that have been dosed with three different concentrations per drug.

Rat hair was collected on the first day of dosing and the last day of dosing from each rat. The black and white hair was separated prior to analysis. The hair samples were combined by the low dose, medium dose, and the high dose for each drug. Three rats were dosed with saline and served as the control group. Once the hair was collected, the samples were washed and digested in hydrochloric acid (0.5M) overnight at 45°C. The samples were then neutralized in sodium hydroxide (0.5M) and buffered in ammonium buffer (0.5 M, pH 8.0). The samples were extracted by SPE using a C-18 cartridge. Prior to evaporation, 100µl of 1mg/ml tartaric acid in ethyl acetate was added. The extract was then evaporated and derivatized using pentafluoropropionic acid (PFPA):ethyl acetate (2:1), and subsequently analyzed using GC/MS in selected ion monitoring mode. The method was linear from 0.1ng/ml to 5ng/ml ($r^2 > 0.99$).

The use of SPE followed by derivatization allowed for the simultaneous detection by GC/MS of amphetamine, methamphetamine, mephedrone, methylone, and ketamine. As predicted, the black hair incorporated all drugs to a higher degree than the white hair and a dose-dependent concentration increase was observed in the black hair. This method will be of great use to the field of forensic hair analysis as it shows the use of SPE/GC/MS for the simultaneous extraction and detection of amphetamine-type stimulant analogues and ketamine.

Hair Analysis, Bath Salts, GC/MS

K24 Benefits of Deconvolution Reporting Software in Forensic Toxicology

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After attending this presentation, attendees will understand how the use of Deconvolution Reporting Software (DRS) can improve efficiency in a busy toxicology laboratory.

This presentation will impact the forensic science community by decreasing the time an examiner spends performing routine data analysis and decreases the likelihood that substances are overlooked during labor-intensive manual searches.

Compounds of interest were added to the Agilent® Deconvolution Reporting Software (DRS). An existing validated procedure for the analysis of Basic, Acidic, and Neutral (BAN) drugs was used throughout the study. The method was retention-time locked for mepivacaine, one of three internal standards used in the procedure.

Deconvolution is a process by which ions are extracted from a complex Total Ion Chromatogram (TIC) and helps to

identify compounds even when the target compound signal is at trace levels and/or hidden under co-eluting matrix compounds. The deconvolution reporting software for Gas Chromatography/Mass spectrometry (GC/MS) is designed to target compound analyses which combine data from the Agilent® Mass Selective Detector (MSD) Productivity ChemStation, the National Institute of Standards and Technology (NIST) Automated Mass Spectral Deconvolution and Identification Software (AMDIS), and the NIST 2008 Mass Spectral Search Program (NIST 08) into one simple report.

Over 100 of the most commonly reported drugs and metabolites were added to the DRS library. For the purpose of this evaluation, only drugs and drug metabolites were added that do not need special treatment, such as formation of a derivative. To evaluate the time taken to identify substances using the traditional (manual) and DRS approach, the same examiner evaluated a positive control blood sample containing 26 drugs. Manually, this identification process took 10-15 minutes. By comparison, the time to scan the TIC and produce the deconvolution report was less than a minute. The NIST library was searched for the components that were found in the AMDIS target library. Retention times, retention time differences, Chemical Abstracts Service (CAS) numbers, and percentage matches are also included in this report for each compound that was identified.

To determine the concordance of results between manual and automated approaches, a retrospective side-by-side comparison of 20 previously analyzed toxicology cases was performed. In three cases, the DRS identified additional substances that were not originally reported: fentanyl, diphenhydramine, temazepam, and a trace level of phenobarbital. In all three cases, these additional substances met the reporting criteria of the laboratory. In the 17 remaining cases, the DRS report was in agreement with the reported results or tentatively identified a substance that did not meet reporting criteria due to the quality of the library match. In addition to decreasing analysis time and improving efficiency, the DRS approach performs very well on complex biological matrices that may contain significant coextractives and interferences. These findings and the utility of DRS in routine forensic toxicology casework will be discussed.

DRS, GC/MS, AMDIS

K25 Detection of Drugs in Postmortem Blood and Skeletal Tissue (Bone)

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After attending this presentation, attendees will have a better understanding of the utilization and interpretive value of the detection of drugs in postmortem skeletal tissue (bone).

This presentation will impact the forensic community by informing forensic professionals how to utilize alternate matrices such as bone when traditional specimens such as blood, body

fluids, and tissues are not available for toxicological analysis.

A recent multiple homicide case where only skeletonized and extremely putrefied fluids were available prompted the laboratory to establish a standardized approach for the analysis of bone in future cases. To do this, bones from non-decomposed specimens were evaluated. Cardiac and Femoral Blood (FB), along with routine postmortem specimens (iliac crest or vertebrae) were collected in 20 cases. Specimens were analyzed using a 13-panel Enzyme-Linked Immunosorbent Assay (ELISA), base and acidic/neutral, Enzyme Multiplied Immunoassay Technique (EMIT), acetaminophen, and salicylate analyses. Confirmations/quantitations were performed using gas chromatography/mass spectrometry.

Bones were stored at -20°C for three years prior to analysis. Cleaned bones were rinsed with deionized (DI) water, dried for 48 hours, manually crushed, and ground in a commercial grinder. Bone (3g) was added to 10mL methanol, vortexed 1 minute, incubated 48hrs at room temperature, and rotary extracted for 8hrs. Methanol supernatants were recovered by centrifugation, evaporated to near dryness, combined with 100µl of 0.1% methanolic HCl. Phosphate Buffer (PB: 2mL, 100mM, pH 6) and 4mL acetonitrile: methanol (2:1 v/v ratio) were added to evaporated extracts. Vortexed extracts were stored at -20°C for 12 hours, then centrifuged 5 minutes. Supernatants were reduced to 2mL by evaporation, and diluted to 5mL with PB. Bone extracts were analyzed in the same manner as blood. Drug-free bones from postmortem blood screening served as negative controls (n=3).

Opioids were detected in 8/9 cases, which screened positive by ELISA. In two heroin cases, only codeine was detected, 6.6ng/g and 15ng/mL, bone and FB, respectively in one case, no opiates were present in the other. Two cases contained hydrocodone, 36 and 8.3ng/g bone; 76 and 44ng/mL FB. Dihydrocodeine was detected in both FB samples, but not in bone. Oxycodone was detected in three cases at 161, 25, and 83.5ng/g, bone and 520, 65, and 920ng/mL FB. Oxymorphone was present in one bone specimen and in all three FB. Fentanyl, methadone, and propoxyphene were detected in one case each: (4.17ng/g, bone, 8.7ng/mL, FB), (217ng/g, bone, 530ng/mL, FB), (1.35ng/g bone, and 2.9mg/L FB), respectively. EDDP was present in FB, 220ng/mL, but not in bone. Norpropoxyphene was present in both matrices.

Methylenedioxypyrovalerone (MDPV) and N-benzylpiperazine (BZP) were present in individual cases in FB, urine, and bone. Synthetic cannabinoids JWH-18 and JWH-250 were detected in one case, bone, 0.33 and 1.4ng/mL and FB, 0.48, 4.6ng/mL, respectively, with AM-2201 and RCS-4 being found in bone only, synthetic cannabinoid analysis performed by NMS Labs. Cocaine/metabolites were present in four cases: benzoylecgonine, <20, 62, 258, and 512ng/g for bone, and 88, 96, 830, and >2000ng/mL for FB. EME was in two bone samples, anhydroecgonine methylester (AEME) was present in one bone sample and not detected in FB. Cocaine was present in one case at 58.4ng/g, bone, 278ng/ml, FB.

Nordiazepam was detected at 67.8ng/g, bone, qualitatively present in FB. Clonazepam was detected in two FB samples, but not in the corresponding bone. Cyclobenzaprine was present in one specimen, 183ng/g, bone, 14ng/mL, FB. Venlafaxine was present in one case, 768ng/g, bone, 0.69mg/L, blood. Norvenlafaxine and lidocaine were present in both matrices. Dextromethorphan was present in bone, 968ng/g and >2000ng/L for FB, in an overdose case. Diphenhydramine was present in two FB, 1.45 and 0.075mg/L, but was only seen in the first at 300ng/g, bone. Cotinine correlated in 6/7 cases with FB and urine. Nicotine and caffeine were not found in bone, both detected in FB. Doxylamine, promethazine, and ibuprofen were also found in both bone and FB.

In these cases, most drug findings in blood were also

detected in bone. In no cases were drug concentrations higher in bone than in blood. These findings demonstrate that bone may be a useful matrix to document drug usage in postmortem cases when traditional specimens are unavailable.

Bone, Skeletal Tissue, Alternate Specimens

K26 Optimizing the Best pH (3 or 7) for the Extraction of Cannabinoids From Whole Blood Samples in Drugs and Driving Cases

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After attending this presentation, attendees will be able to choose the most efficient pH value for extracting cannabinoids (tetrahydrocannabinol (THC), 11-hydroxytetrahydrocannabinol (THC-OH), tetrahydrocannabinol-carboxylic acid (THCA), cannabidiol (CBD), and cannabinol (CBN)) from whole blood samples employing available Solid Phase Extraction (SPE) cartridges and liquid chromatography-tandem mass spectrometry.

This presentation will impact the forensic science community by offering analysts operating in forensic facilities information regarding the extraction and analysis of popular cannabinoids encountered in drugs and driving casework using solid phase extraction and liquid chromatography-tandem mass spectrometry.

Method: Samples of whole blood ((1mL) calibrators, controls, and case samples) were spiked with THC, THC-OH, THCA, CBD, and CBN (plus THC-d3, THC-OH-d3, THCA-d3, and CBD-d3). Samples were precipitated with 2mL of acetonitrile. The acetonitrile was removed and evaporated to 500µL, then diluted with either A (5mL of 0.1M aqueous hydrochloric (pH 3)) or B (5mL of 0.1M phosphate buffer (pH 7)). This comparison study was performed using validation guidelines at Massachusetts State Police Crime Laboratory.

Extraction of the A samples was performed on commercially available SPE columns (C8/Aminopropyl). These SPE columns were conditioned with methanol, deionized (DI) water, and 0.1M aqueous hydrochloric acid (3mL, 3mL, 1mL, respectively) and the samples loaded onto the SPE cartridges at 1mL/minute. The SPE columns were washed with DI water and 30/70 acetonitrile-0.1M HCl (3mL) before drying (10 minutes), after which the cannabinoids (and internal standards) were eluted with 3mL of a hexane/ethyl acetate/acetic acid (49:49:2) mixture. The eluates were evaporated to dryness under nitrogen and dissolved 100µL of methanol for analysis. The B samples were extracted on the same type of SPE columns pre-conditioned with methanol, DI water, and 0.1M phosphate buffer (pH 7) (3mL, 3mL, 1mL, respectively) after which the pH 7 buffered samples were loaded onto the SPE columns at 1mL/minute. The SPE columns were washed with pH 7 phosphate buffer (3mL) before drying. The cannabinoids were eluted with 3mL of the same elution solvent. The eluates were evaporated to dryness under nitrogen before being dissolved in 100µL of methanol for analysis.

Liquid chromatography was performed in gradient mode employing a 50mm x 2.1mm (5µm) C₁₈ analytical column and a mobile phase consisting of acetonitrile and 0.1% aqueous formic acid. The gradient was programmed to run from 5% to 90% acetonitrile in 4.0 minutes and then back to 5% for re-injection. The total run time for each analysis was less than 5 minutes.

Tandem mass spectrometry was performed in positive/negative Multiple Reaction Monitoring (MRM) modes. The following transitions were monitored (quantification transition ions

underlined): THC m/z: 315.3 to 193.2, 123.2; THC-d₃ m/z 318.3 to 196.2, 123.1; THC-OH: m/z 331.1 to 193.1, 201.1; THC-OH-d₃: m/z 334.3 to 196.2, 133.1; THCA: m/z 343.3 to 299.1, 245.1; THCA-d₃: m/z 346.1 to 302.1, 245.1; CBD: m/z 313.2 to 245.2, 245.2; CBD-d₃ m/z: 316.2 to 247.8, 182.1; CBN: m/z 311.3 to 223.2, 178.1, respectively. In this presentation, representative chromatograms are shown to illustrate the efficiency of the chromatography and analysis of THC and related cannabinoids from 20 (completed) drugs and driving cases.

Results: The limits of detection/quantification for the SPE method were determined to be 0.5 ng/mL and 1.0ng/mL, respectively for all the cannabinoids. Both pH values (pH 3 and pH 7) methods were found to be linear from 1ng/mL to 100ng/mL ($r^2 > 0.999$). Data is presented to show that recoveries of all the cannabinoids were found to be greater than 90%. Interday and intraday analysis were found to <7% and <10%, respectively. Matrix effects were determined to be <5% for both types of extraction.

Conclusion: Extraction of cannabinoids at pH 3 demonstrates the efficient use of hydrophobic interactions, while extraction at pH 7 demonstrates the use of both hydrophobic and anion exchange interactions leading to better sensitivity and selectivity in the retention of the cannabinoids on mixed mode SPE columns leading to better evaluation of results. The procedure of extracting cannabinoids from whole blood samples using SPE at pH 7 offers better recovery values (>95%) than pH 3 (>90%). This information should assist analytical toxicologists when choosing an efficient method for the analysis of popular cannabinoids in whole blood samples.

Cannabinoids, Blood, SPE

K27 Enzyme Linked Immunosorbent Assay (ELISA) for Detection of Use of the Synthetic Cannabinoid Agonists UR-144 and XLR-11 in Human Urine

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After attending this presentation, attendees will be able to describe the method validation parameters used to assess this ELISA kit as well as the performance characteristics established for each parameter during the validation. Attendees will also be able to evaluate the applicability of the UR-144/XLR-11 Direct ELISA kit for screening large-scale populations.

This presentation will impact the forensic science community by providing information about a novel validated ELISA and its performance designed to detect two of the currently most prevalent synthetic cannabinoids in urine.

The purpose of this project was to develop and validate an ELISA designed to test for metabolites of the synthetic cannabinoid UR-144 and its fluorinated homolog XLR-11 in urine.

The dynamic synthetic cannabinoid drug market has created the need to continually revisit and update approaches for screening for use of these drugs in urine. Prior to use for forensic purposes, these approaches need to be thoroughly validated. The current federal regulations regarding these compounds and the inability to detect synthetic cannabinoids on traditional marijuana immunoassay tests have created the need for sensitive cost-effective assays capable of screening large-scale populations. As

recently as 2012, tetramethylcyclopropylindoles, including UR-144 and its closely related analog XLR-11 began to appear on the Novel Psychoactive Substance (NPS) market. Like other synthetic cannabinoids, UR-144 and XLR-11 act on the cannabinoid receptors (CB1 and CB2) in the central and peripheral nervous system to elicit effects similar to delta-9-tetrahydrocannabinol (THC).

In this study, the UR-144/XLR-11 ELISA kit was validated for use on human urine samples, although it is also suitable for use on other matrices. The principle of the ELISA kit was based on the competitive binding of the analyte and a UR-144 pentanoic acid metabolite-peroxidase conjugate, to a proprietary antibody coated on the 96-well microplate. The assay was calibrated at 5ng/mL with the pentanoic acid metabolite of UR-144. The cut-off of 5ng/mL was selected to reflect the concentrations of metabolites encountered in actual subject samples from a large screening population tested in-house. The performance of the assay below the cutoff concentration of 5ng/mL was not challenged.

The UR-144/XLR-11 assay has no significant cross-reactivity with most other commonly encountered synthetic cannabinoids and their metabolites, such as JWH-018, AM-2201, JWH-210, and others. However, the assay does cross-react with UR-144-5-OH metabolite (100%), UR-144-4-OH metabolite (50%), and XLR-11-4-OH metabolite (50%) in addition to UR-144 pentanoic acid, which is used as the calibrator. Besides a broad range of synthetic cannabinoids and their metabolites, a range of therapeutic drugs and several commonly used drugs of abuse including benzoylecgonine, cocaine, codeine, EDDP (a metabolite of methadone), MDMA, methamphetamine, methadone, morphine, and PCP in addition to THC, 11-nor-9-carboxy-THC, 11-hydroxy-THC, and cannabidiol were evaluated to determine potential interference with the assay. None produced any positive results at a concentration of 20,000ng/mL.

The long-term stability of the kit is currently under investigation. To date, the assay appears to be stable and performed acceptably after 4.3 months. With respect to intraplate and interplate precision, the UR-144/XLR-11 kit was precise with a coefficient of variation of less than 15% for the negative (3.75ng/mL), positive (6.25ng/mL), and cutoff (5.0ng/mL) controls. Both carryover and plate drift were examined using the UR-144 pentanoic acid calibrator. The assay was free from carryover at a concentration of 1,000ng/mL and no plate drift was observed.

One hundred blind controls were prepared for screening by the assay. Forty were negative controls, verified by LC/MS/MS, to which no drug or metabolite was added. Sixty other positive controls that contained UR-144-4-OH, UR-144-5-OH, and UR-144 pentanoic acid were spiked at varying concentrations ranging from 0-100ng/mL. The assay yielded 40 true negatives and 60 true positives, giving an overall sensitivity, specificity, and accuracy of 100% for the assay.

The rapidly changing synthetic cannabinoid market and continuous regulation of these compounds, including UR-144 and XLR-11, has created a need for rapid screening techniques capable of handling high volumes of samples. Using the UR-144 Direct ELISA assay kit, screening urine samples is both highly cost effective, specific, and had appropriate sensitivity for actual patient populations.

Synthetic Cannabinoids, ELISA Kits, Urine Drug Screening

K28 SOFT/AAFS Drugs and Driving Special Scientific Session

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After attending these presentations, attendees will have a greater understanding of the prevalence of drug use in drivers, challenges Driving Under the Influence of Drugs (DUID) cases pose for forensic laboratories, and how those challenges effect law enforcement and prosecution.

These presentations, focused on DUID, will impact the forensic science community's understanding of the extent of DUID cases, providing specific research findings related to several drugs' effects on the skills required to safely operate a motor vehicle and the far-reaching implications for law enforcement and attorneys.

Introduction: DUID cases pose unique challenges not only to forensic laboratories, but to law enforcement and the legal community as well. Forensic laboratories can vary significantly in their resources, scope of testing, and, in some cases, can be restricted by local legislation. This special session will discuss the efforts of the National Safety Council's Alcohol, Drugs and Impairment Division to provide guidelines for DUID laboratories and the impact those guidelines have on law enforcement and prosecution of DUID cases. Specific examples of impairment that result from driving under the influence of marijuana in a *per se* legislation state as well as presentations regarding phenobarbital and synthetic cathinones impairment will be presented.

The Importance of Standardization for DUID Laboratories: Forensic laboratories involved in DUID casework utilize a wide variety of resources which leads to difficulty in standardization of testing. The National Safety Council's Alcohol, Drugs and Impairment Division appointed a subcommittee to address the similarities and differences across various DUID laboratories. The process included surveys from each state's Traffic Safety Resource Prosecutors (TSRP), Drug Recognition Expert (DRE) State Coordinators, and forensic laboratories performing DUID testing. The subcommittee examined the surveys and, subsequently, established guidelines for the appropriate scope of testing for these laboratories. The guidelines set forth will be a useful tool for laboratories to use to provide the appropriate support for law enforcement arrests involving individuals driving under the influence of drugs.

Re-examining the "Three-Legged Stool" Approach to Detering Drugged Driving: As the Drug Evaluation and Classification (DEC) Program and Advanced Roadside Impaired Driving Enforcement (ARIDE) training continues to increase nationally, more suspected drug-impaired drivers are being arrested on our nation's roadways. With the increased number of officers being trained to detect drug impairment, additional workloads are being placed on toxicologists and forensic laboratories to support law enforcement opinions and to report toxicology findings in a timely manner to assist in the prosecution of these cases. An important part of the DRE training is the understanding of the "three-legged stool" concept which includes the DRE opinion, toxicology, and prosecution. This presentation will emphasize the three important "legs" that are needed to support the DEC Program and the efforts to deter drug-impaired driving.

Lab Analysis and the Law: The Impact of *Bullcoming v. New Mexico* on Forensic Science Testimony: This presentation will provide a prosecutor's overview of the impact on forensic analysis testimony versus basic maintenance or procedural testimony following the U.S. Supreme Court's 2011 ruling in *Bullcoming*. Although the factual distinction has succeeded in most

Washington courts, some jurisdictions continue to require a specific lab analyst and/or breath test technician to testify even regarding routine procedural processes.

Marijuana Impaired Driving in a Marijuana-Legal State: In December 2012, the possession and private use of marijuana became legal in the state of Washington. At the same time, a *per se* level of 5ng/mL of delta-9-THC in blood came into effect. In December 2013, marijuana products will be commercially available to the public via state-licensed facilities. This presentation will provide an overview of suspected driving under the influence cases involving marijuana pre and post the legalization of marijuana in Washington.

Butalbital and Driving Impairment: Butalbital (Fiorinal®) is a barbiturate commonly prescribed for the treatment of tension headaches and migraine. Butalbital has been reported to be the most commonly encountered barbiturate in DUID cases. Butalbital has common Central Nervous System (CNS) depressant properties, with side effects including sedation, drowsiness, and feelings of intoxication which can contribute to driving impairment. Twenty-six DUID cases from the state of Washington are reviewed with results from field sobriety tests and toxicological findings included. Butalbital whole blood concentrations ranged from 1.0 to 30.2mg/L, with a mean and median of 16.0mg/L. General impairment indicators in these cases included horizontal and vertical nystagmus, lack of convergence, poor motor coordination, and balance and speech problems which are common to CNS depressant intoxication, similar to that associated with alcohol. These findings indicate the importance of toxicological testing for butalbital in cases where CNS depressants are indicated.

Synthetic Cathinones and Driving Performance: Designer drugs have proven challenging for forensic laboratories both analytically and during interpretation of laboratory results. Currently, there is very little in the scientific literature to document cases of driving under the influence of synthetic cathinones. This presentation will closely examine the impairment documented by law enforcement during recent DUID investigations and the related toxicology results. Challenges including limited availability of reference materials, method validation, and the ability to predict the next synthetic cathinone will also be discussed.

Drugs and Driving, Impairment, Forensic Toxicology

K29 Δ-9-Tetrahydrocannabinol (THC) Concentrations in Drivers Testing Positive for Marijuana Use and Consequences for the Effectiveness of a THC *Per Se* Law

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After attending this presentation, attendees will be able to describe patterns of combined drug and alcohol use in drivers testing positive for marijuana use, and assess the likely effectiveness of *per se* thresholds for delta-9-tetrahydrocannabinol (THC) in blood samples in Driving Under the Influence of Drugs (DUID) cases.

This presentation will impact the forensic science community by providing information about: (1) the impact of setting arbitrary *per se* thresholds for drugs like THC with rapidly changing pharmacokinetic profiles; and (2) on the effectiveness of *per se* laws governing DUID.

This study was undertaken to assess the typical range of THC concentrations in blood samples submitted in DUID investigations relative to the *per se* concentration thresholds in

various U.S. states.

These results were obtained from blood samples submitted to the laboratory for confirmatory testing for marijuana use in DUID cases between August 2009 and June 2013, after having screened positive by a cannabinoid Enzyme-Linked Immunosorbent Assay (ELISA). Cases were analyzed for delta-9-tetrahydrocannabinol (THC, cut off 1ng/mL), 11-OH-delta-9-tetrahydrocannabinol (THC-OH, cut-off 5ng/mL), and 11-carboxy-delta-9-tetrahydrocannabinol (THC-COOH, cutoff 5ng/mL), using three-dimensional gas chromatography-mass spectrometry.

A total of 3,814 cases tested positive for THC above 1ng/mL, and were additionally tested for alcohol by Headspace Gas Chromatography with Flame Ionization Detection (HS-GC/FID) and for other drug classes (benzodiazepines, opiates, cocaine metabolite, amphetamines, methadone, phencyclidine, barbiturates, and propoxyphene) by ELISA. These cases were categorized as follows: (1) positive for THC with other drugs and alcohol present (>0.01) (n=558; 14.6%); (2) positive for THC without other drugs but with alcohol present (>0.01) (n=1,625; 42.6%); (3) positive for THC with other drugs detected but no alcohol (n=619; 16.2%); and, (4) positive for THC with no other drugs or alcohol present (n=1,012; 26.5%). In summary, 30.8% were positive for other drug use in addition to THC, while 57.2% were positive for alcohol use in addition to THC.

Within these groups, cases were evaluated as to whether the THC concentration exceeded certain thresholds, specifically, the 2ng/mL *per se* threshold applied in Ohio and Nevada, and the 5ng/mL threshold applied in Colorado and Washington state.

Table 1: Distribution of THC Concentration values at key *per se* thresholds by alcohol/other drug status. All cases were positive for THC at a concentration of 1.0 or greater.

	n	Median THC concentration (ng/mL)	1 – 1.9ng/mL (Below <i>per se</i> threshold in OH, NV)	1-4.9ng/mL (Below <i>per se</i> threshold in WA/CO)
All THC Positive cases with Alcohol and Drug results	3,814	3.8	24.2%	62.8%
THC +; Alcohol +; Other Drug +	558	2.8	33.5%	73.6%
THC +; Alcohol +; Other Drug -	1625	3.1	27.5%	70.1%
THC +; Alcohol -; Other Drug +	619	3.7	25.7%	61.7%
THC +; Alcohol -; Other Drug -	1012	5.6	12.8%	45.6%

The data demonstrate that a significant proportion of all drivers tested (24.2%) had blood THC concentrations below the *per se* thresholds in Ohio and Nevada, while 62.8% had concentrations below the *per se* thresholds in Washington and Colorado. Broken down further by the drivers' alcohol and other drug use status, cases where THC was the only drug present and the most likely cause for the observed impairment had the smallest proportion of drivers under the 5ng/mL *per se* threshold (45.6%) and the highest median THC concentration (5.6ng/mL). Cases with both alcohol and other drugs present where not all the observed impairment can be attributed to THC, had the lowest median THC concentration (2.8ng/mL) and the highest proportion of drivers who would escape the 5ng/mL *per se* prosecution at 73.6%. Cases with THC and alcohol only, or cases with THC and other drugs only, fell in between these two extremes.

The application of 5ng/mL *per se* laws for THC in drivers risks excluding a high number of drivers whose blood THC concentrations will fall below the *per se* threshold during the 1-2 hours it takes to collect a blood sample following the stop, investigation, and arrest.

Officials in jurisdictions with 5ng/mL *per se* or presumptive concentration thresholds in their statutes for THC should give significant consideration to objective evidence of impairment and presence of symptoms associated with marijuana use, rather than relying solely on blood THC concentrations that fall below these arbitrary thresholds when determining whether a subject is under the influence as a result of their marijuana use.

Marijuana, DUID, Per Se Laws

K30 Breath Ethanol Concentration in Obese and Underweight Humans

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After attending this presentation, attendees will learn about the effect of obesity on the breath ethanol concentration generated from dosing humans and gain insight to improve applications with this concept.

This presentation will impact the forensic science community by providing data to assist with estimating an ethanol concentration in obese and underweight humans.

Ethanol is hydrophilic and distributes at equilibrium in body fluids and tissues in proportion to their water content and has low solubility in fat (adipose) tissue. A subject with a higher proportion of body fat and concomitant lower proportion of body water is hypothesized to have a higher relative ethanol concentration in blood or breath; conversely a lower proportion of body fat and higher proportion of body water is hypothesized to have a lower relative ethanol concentration. Methods to estimate a blood ethanol concentration in humans derived from experimental data exist; however, their accuracy has not been reliably addressed with the obese (Body Mass Index (BMI) ≥ 30) and underweight (BMI < 18.5). The prevalence of obesity has significantly increased and is now estimated by the Centers for Disease Control and Prevention (CDC) at 35% of American adults.¹ Breath samples were collected about every 15 minutes from 806 forensic human subjects (86.6% male drivers) following a controlled drinking test and analyzed using a Breathalyzer™ 900/900A or Intoxilyzer® 5000 instrument. The Ethanol Concentration per Dose (EC/D) was calculated by dividing the y-intercept of breath ethanol (alcohol) concentration versus time by the total dose of ethanol administered. The relative body-paired EC/D was determined for obese (17.3%) and underweight (1.7%) subjects that could subsequently be paired with control subjects in the target (ideal) BMI range with the same gender, age, and similar height (up to ± 3 cm for males, ± 5 cm for females). The relative body-paired ethanol concentration per dose (bp-EC/D) was calculated from the ratio of subject EC/D multiplied by the inverse ratio of their body masses, with results summarized below:

		bp-EC/D			BMI: Condition Subjects		BMI: Control Subjects	
Condition	Sex	Median	Range	N	Median	Range	Median	Range
Obese	M	1.113	0.642 to 1.711	65	32.2	30.0 to 42.0	23.5	19.7 to 24.9
Obese	F	1.154	1.049 to 1.155	3	34.9	30.2 to 46.7	21.8	20.4 to 23.8
Underweight	M	0.887	0.758 to 1.012	3	18.2	17.7 to 18.3	22.5	21.6 to 23.2
Underweight	F	0.883	0.798 to 1.015	6	17.6	16.1 to 18.4	21.4	18.5 to 22.0

These results from the body-paired groups agree (i.e., are not inconsistent) with the hypothesis for both gender (male, female) and body conditions (obese, underweight) studied. The magnitude of the effect of obesity on the ethanol concentration is similar to that generally attributed to gender. Estimation of the EC/D using equations derived by Watson et al., adjusted for mass-per-volume units of measure by the average density of blood (1.055g/mL), provided the best overall agreement with breath ethanol test results for the obese and underweight humans studied.² The median relative ethanol concentration per dose (experimental/theoretical) from the adjusted Watson approach was similar and close to unity for the four groups: (1) 0.984 (range: 0.724 to 1.323, N=128) for obese males; (2) 0.957 (range: 0.721 to 1.206, N=12) for obese females; (3) 0.988 (range: 0.844 to 1.066, N=6) for underweight males; and, (4) 0.999 (range: 0.845 to 1.060, N=8) for underweight females.

References:

1. C.L. Ogden, M.D. Carroll, B.K. Kit & K.M. Flegal, "Prevalence of Obesity in the United States, 2009-2010", NCHS Data Brief 82 (Jan. 2012). Online: Centers for Disease Control and Prevention, <http://www.cdc.gov/nchs/data/databriefs/db82.pdf>.
2. P.E. Watson, I.D. Watson & R.D. Blatt, "Prediction of Blood Alcohol Concentrations in Human Subjects" J. Stud. Alc. 42(7), 547-556 (1981).

Ethanol, Obesity, Concentration

K31 Driving Under the Influence of the Synthetic Cannabinoid Receptor Agonist XLR-11

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After attending this presentation, attendees will become aware of a Driving Under the Influence of Drugs (DUID) case involving the synthetic cannabinoid receptor agonist XLR-11 including driving pattern, driver's demeanor and behavior, performance on field sobriety tests, and analytical findings.

The presentation will impact the forensic science community by reporting for the first time a complete case report of a driver who was found driving under the influence of the synthetic cannabinoid receptor agonist XLR-11.

A 22-year-old White man was involved in a traffic collision when the truck he was driving rear-ended a passenger vehicle lawfully stopped at a busy San Francisco intersection. Witnesses at the scene reported that the driver had a blank stare on his face and kept looking straight ahead. He was reportedly "very high." His speech was described as mellow and his voice was barely audible; he appeared rigid and his muscle tone was described as tense. A police Drug Recognition Expert (DRE) performed an evaluation of the driver and found, among other signs, low body temperature, rigid muscle tone, normal pulse, lack of horizontal gaze nystagmus, lack of vertical gaze nystagmus, non-convergence

of the eyes, dilated pupil size, and normal pupillary reaction to light. The DRE concluded that this driver was driving while under the influence of cannabis. A standard toxicology DUID protocol was employed utilizing: (1) headspace gas chromatography equipped with flame ionization detection for ethanol and related volatiles; (2) Enzyme Linked Immunosorbent Assay (ELISA); and, (3) Gas Chromatography coupled with Mass Spectrometry (GC/MS) for amphetamines, barbiturates, benzodiazepines, cannabinoids, cocaine, methadone, phencyclidine, opiates/opioids, and more than 100 other drugs and metabolites, but produced completely negative results. Additional screening of the whole blood specimen was then undertaken by a reference analytical laboratory for synthetic cannabinoid receptor agonists by Liquid Chromatography-Tandem Mass Spectrometry (LC/MS/MS) on a Waters ACQUITY® Ultra Performance Liquid Chromatographic (UPLC) system coupled with a Tandem Quadrupole mass spectrometric Detector (TQD). The analytical column utilized was an ACQUITY® UPLC BEH C18 reversed-phase column (1.7µm, 2.1mm x 100mm) which has a very wide usable pH range (pH 1-12) and is suitable for UPLC separations due to its trifunctionally-bonded Ethylene Bridged Hybrid (BEH) particles. The mobile phase was a mixture of 0.1% formic acid in deionized water and 0.1% formic acid in methanol. The injection volume was 20µL. Data analysis was performed on the MassLynx™ MS Software platform. The synthetic cannabinoid receptor agonist XLR-11 was identified by monitoring the MS/MS ion transitions 330.3 → 125.2 and 330.3 → 232.3. XLR-11 was quantified in a fresh aliquot of blood using calibration curves made by spiking drug-free blood at 0.10, 0.20, 1.0, 4.0, 10.0, and 20.0ng/mL. XLR-11 was quantified in this driver's whole blood at a concentration of 1.34ng/mL. Although other synthetic cannabinoid receptor agonists have previously been encountered by the Division in postmortem toxicology cases and drug-facilitated sexual assaults, this is the first documented case involving a San Francisco driver driving under the influence of the synthetic cannabinoid receptor agonist XLR-11.

DUID, Spice XLR-11, DRE

K32 The Scientific Working Group in Toxicology (SWGTOX): An Update on Standards Development

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After attending this presentation, attendees will learn the current status of standards development within the Scientific Working Group in Toxicology (SWGTOX). Attendees will understand how they can actively participate in the standards development process.

This presentation will impact the forensic science community by delineating the work of SWGTOX and how it

potentially impacts the day-to-day activities within forensic toxicology laboratories within the United States.

The Scientific Working Group in Toxicology was established in October 2009. Its mission is to develop and disseminate consensus standards for the practice of forensic toxicology in the United States. The main areas of focus include:

- Standards, practice, protocols including QA and QC
- Education and training requirements
- Accreditation
- Certification
- Develop a Code of Professional Conduct for forensic toxicologists and laboratories
- Identify general areas of Research and Development (R&D) needs
- Promote public awareness of the field of forensic toxicology

The scope of SWGTOX activities concerns toxicological matters including postmortem toxicology, human performance toxicology, workplace drug testing, medicolegal and criminal investigations, and court-mandated testing.

To this end, members of SWGTOX have worked diligently within committees, subcommittees, and task groups to complete a number of standards. Other standards are on-going and in various stages of completion. It is important for the entire forensic toxicological community to be aware of SWGTOX activities and understand how each can play a role in the promulgation of standards. This presentation is an update of SWGTOX activities for the community. Executive Committee members of SWGTOX will present updates on specific accomplishments and current standards being developed.

SWGTOX, Standards, Participation

K33 Analysis of Alpha PVP, MePPP, Naphyrone, and Seven Related Pyrrolidinophenones in Blood by LC/TOF and LC/MS/MS

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After attending this presentation, attendees will be able to describe the characteristics and effects of pyrrolidinophenones, optimum methods for their analysis in blood using a screen and confirmation combination of Liquid-Chromatography/Time-of-Flight (LC/TOF) and Liquid Chromatography/Electrospray Ionization/Tandem Mass Spectrometry (HPLC/ESI/MS/MS), and to take into account limitations on its analysis in interpreting results.

This presentation will impact the forensic science community by describing a comprehensive assay for the detection of this emerging class of New Psychoactive Substances (NPS).

Pyrrolidinophenones are an emerging group of designer drugs that are appearing in NPS. Compounds in this class are characterized by a propiophenone backbone attached to a pyrrolidine group, with various substituents on the aromatic ring and modifications at the alkyl chain producing the related compounds. One of the most popular compounds of this group that is being recreationally abused is alpha-pyrrolidinopentiophenone (alpha-PVP), which is a designer stimulant that can cause many adverse effects, including acute psychosis with delusions as well as other medical issues including cardiovascular problems, seizures, and hyperthermia. Other compounds of the pyrrolidinophenone class

have been seen on the market. Alpha-PVP and MePPP have both been reported in synthetic drug material and alpha-PVP, MDPPP, and MPHP have all been reported in case histories of individuals using "bath salts" or "legal high" products. Due to the emerging nature of this group of compounds, it is necessary to develop testing for the detection of pyrrolidinophenones in forensic toxicology.

This method was developed for the detection of ten different compounds in the pyrrolidinophenone class: alpha PVP, pyrovalerone, naphyrone, alpha-PBP, alpha-PPP, MePPP, MDPPP, MOPPP, MPBP, and MPHP. Since many of the pyrrolidinophenones have similar molecular structures, weights, and formulas, it was important to develop a sensitive and reliable method for the determination of these compounds in biological matrices for forensic toxicology.

Pyrrolidinophenones were extracted from whole blood by a single step liquid-liquid extraction. Samples are extracted with a 0.1M borax buffer (pH 10.4) and purified by liquid-liquid partition with a lamotrigine extraction solvent (30% ethyl acetate in n-butyl chloride). Spiked samples were analyzed by LC/TOF mass spectrometry for screening purposes. For the LC/TOF, the mobile phases consisted of 0.05% formic acid in 5mM ammonium formate and 0.05% formic acid and the analysis was performed on a ZORBAX® Eclipse Plus C18 Rapid Resolution HT 3.0 x 100mm column. Target compounds were detected and reported from accurate scan data using MassHunter Qualitative and Personal Compound Database and Library (PCDL) software. Retention time data was obtained from blank matrices spiked at a cutoff concentration with reference material for each drug. A confirmatory technique for biological matrices was also developed using HPLC/ESI/MS/MS. Infusions of each analyte were performed to determine daughter ions; transitions had to be carefully selected due to some compounds having the same molecular mass and almost identical retention times. HPLC conditions for the LC/MS/MS method included ammonium formate vs methanol at 0.40mL/min on an ACQUITY® UPLC™ BEH C18 2.1 x 100mm column. A serial dilution was evaluated for a calibration curve; the final calibration range is 5-500ng/mL. Characterization experiments regarding linearity, matrix-matching, and recovery were successfully completed. This panel separates and identifies ten compounds that have similar structures, molecular weights, and retention times.

This research was the first documented attempt to develop a comprehensive panel for pyrrolidinophenones, which are increasing in popularity for abuse in the new designer drug movement. This panel was developed for identifying these compounds in biological matrices for forensic toxicological analysis. These findings suggest that the combination of LC/TOF and LC/MS/MS provides a valuable approach to the sensitive and specific analytical identification and measurement of pyrrolidinophenones in forensic casework.

Pyrrolidinophenone, Novel Psychoactive Substances, Toxicology

K34 Stability of the Synthetic Cathinones: Implications for Forensic Toxicology

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After attending this presentation, attendees will understand the challenges associated with the stability of the synthetic cathinones and the implications for forensic toxicology analysis and interpretation.

This presentation will impact the forensic community by increasing awareness pertaining to the stability of the synthetic cathinones and the need to consider this factor during analytical and interpretive deliberations.

Synthetic cathinones are an important class of designer drug. The widespread attention and publicity associated with these psychostimulants have resulted in numerous legislative actions at the state and federal level throughout the United States and elsewhere. These amphetamine-like drugs are characterized by a *beta*-keto functional group. Although the synthetic cathinones share many properties of their phenethylamine counterparts, the presence of the ketone moiety is responsible for a number of unique and distinct differences in terms of chemical behavior and in particular, stability.

A series of synthetic cathinones were evaluated as part of a study to investigate novel derivatives by Gas Chromatography/Mass Spectrometry (GC/MS). During the course of this work, it was evident that several drugs within the class were unstable. Degradation of first-generation cathinones was first documented more than 30 years ago in drug chemistry. More recently, forensic toxicology reports have highlighted apparent instability for some of the newer drugs within this class, but more research is needed.

Degradation was most often characterized by a second peak, or shoulder, on the principal analyte yielding a molecular ion 2 mass units lower than the major analyte peak. Corresponding mass losses on the highly characteristic and often dominant iminium ion were also observed. Earlier studies with methcathinone showed that this minor component arose through thermal oxidation of the 2,3-carbon-carbon bond to yield the 2,3-enamine.¹ Oxidative degradation of these arylaminoketones was observed *in situ*. Even tertiary amines, which are reported to be the most stable of the cathinone species, were subject to degradation. GC conditions were optimized to allow for baseline separation for many of the degradation products. Degradation data and spectra illustrating the characteristic fragmentations and proposed structures will be discussed for 3,4-methylenedioxypyrovalerone (MDPV), 4-methyl- α -pyrrolidinobutiophenone (MPBP), pyrovalerone, 3,4-methylenedioxy- α -pyrrolidinobutiophenone (MDPBP), naphyrone, ethylone, mephedrone, 4-Ethylmethcathinone (4-EMC), methcathinone, methedrone, flephedrone, and methylone.

Stability of the arylaminoketones is an important consideration for forensic toxicology. Stability data and mass spectra for the cathinone degradation products will be presented and discussed within the context of forensic toxicological analysis, selection of appropriate instrumental methods, and implications for the interpretation of results.

Reference:

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Cathinone, Stability, GC/MS

K35 Recent Trends of Designer Drugs in Harris County, Texas

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The goal of this presentation is to provide attendees with relevant information about the trends and prevalence of newly emerging designer drugs such as synthetic cathinones (bath salts), cannabinoids (Spice), and designer psychedelics such as 25I-NBOMe (25I, or N-Bomb). This presentation will also provide a forum to discuss the associated challenges that the forensic community is facing every day in discovery, analysis, and interpretation.

This presentation will impact the forensic science community by raising awareness on rapidly changing trends in the availability and use of different classes of designer drugs and the need for the development of cutting-edge analytical strategies to keep up with them. Liquid Chromatography Time-Of-Flight Mass Spectrometry (LC/TOF/MS) has proven to be a successful screening tool, whereas Liquid Chromatography Tandem Mass Spectrometry (LC/MS/MS) is vital in confirming these newly emerging designer drugs and their metabolites. Examining the trends in new drug use and market availability will also help facilitate the scheduling of illicit drugs by local and federal government officials.

This study's laboratory has identified and confirmed designer drugs in 61 (3%) of cases out of a total of 1,992 cases screened involving Driving While Intoxicated (DWI) suspects, victims of sexual assaults, and death investigations since 2011. The screening and confirmation analysis was done on blood (62%) and urine (38%). The drugs currently included in the screening and confirmation panel are 11 cathinones (mephedrone, methedrone, methylone, butylone, ethylone, methylenedioxypyrovalerone (MDPV), naphyrone, α -pyrrolidinopropiophenone (α -PPP), α -pyrrolidinopentiophenone (α -PVP), pentedrone, and 4'-methyl- α -pyrrolidinopropiophenone), 19 cannabinoids and metabolites (JWH-015, JWH-018, JWH-018 N-5-(hydroxypentyl) metabolite, JWH-018 N-pentanoic acid metabolite, JWH-019, JWH-019 N-(6-hydroxyhexyl) metabolite, JWH-073, JWH-073 N-(4-hydroxybutyl) metabolite, JWH-073 N-butanoic acid metabolite, JWH-081, JWH-122, JWH-200, JWH-210, JWH-250, WIN 55212-2, AM-2201, UR-144, UR-144 N-pentanoic acid metabolite, and XLR11), and 7 designer psychedelics (25I-NBOMe, 25B-NBOMe, 25C-NBOMe, 2C-E, 2C-I, TMA-2, and TFMPP).

Out of 61 total cases, 33 cases were deceased individuals and 28 cases were DWI suspects. Bath salts were detected in 38 (62%) of the cases, synthetic cannabinoids were detected in 18 (29%) of the cases, and designer psychedelics were detected in 5 (8%) of the cases. Among the bath salts, the most frequently detected drug was α -PVP, followed by pentedrone, MDPV, ethylone, methylone, and naphyrone. Since 2011, the appearance of synthetic cannabinoids began with JWH-018 and associated hydroxyl/carboxyl metabolites, followed by AM-2201, then giving way more recently to XLR11 and UR-144. In the class of designer psychedelics, the most frequently detected drugs are in the NBOMe series such as 25I-NBOMe and 25B-NBOMe, followed by TFMPP.

This research has found that bath salts are the major designer drugs detected in deceased individuals. In two of these cases, acute toxicity of MDPV was implicated as the cause of death. The toxic effects of α -PVP have been implicated as a cause of death in ten cases. The toxic effect of pentedrone, either alone or with α -PVP, has also been implicated as a cause of death in several cases. The newly emerging designer psychedelics 25I-NBOMe and 25B-NBOMe have been implicated as a cause of death in

four cases. Although synthetic cannabinoids are a frequent finding in DWI and death investigations, there are no deaths classified under synthetic cannabinoid toxicity. Three cases representative of each class of the designer drugs will be discussed, including investigational scene information about the subject, behavioral and pathological findings, and the comprehensive toxicology results.

Bath Salts, Designer Psychedelics, Synthetic Cannabinoids

K36 Stability of 68 Stimulant/Hallucinogenic Drugs in Biological Samples Under Various Storage Conditions

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After attending this presentation, attendees will be able to use information about the stability of 68 stimulant and hallucinogenic drugs in blood, serum/plasma, and urine matrices under various storage conditions.

This presentation will impact the forensic science community by providing information to allow forensic science practitioners to choose the optimal storage conditions to ensure the least degradation of a wide variety of stimulant and hallucinogenic compounds, including several synthetic cathinones and designer drugs.

Synthetic stimulant and hallucinogenic drugs have seen highly increased popularity in the United States since 2009. Drugs in the phenethylamine, cathinone, pyrrolidophenone, tryptamine, and miscellaneous other categories present analytical challenges to forensic toxicologists in the development and validation of screening and confirmation methods. Due to the potential for degradation, it further complicates a laboratory's ability to successfully confirm the presence of these compounds in biological samples, and the client's ability to interpret the analytical findings. Knowing the stability of some of the most prominent drugs in this category is essential to ensuring that accurate results are obtained and reported. The objective of this presentation is to demonstrate the stability for each of 68 compounds of interest including novel and established drugs with stimulant and hallucinogenic effects in various matrices and storage conditions. This will allow attendees to use the most appropriate conditions to maintain drug stability in blood, serum/plasma, and urine.

Stability controls were prepared containing mixtures of the target analytes in blood, serum, and urine. All controls were analyzed in triplicate at room temperature (dark and light), refrigerated, and frozen at days 1, 3, 7, 14, and 30. Compounds of interest included: cocaine; 2C-H; 3-FMC; flephedrone; psilocin; 4-MEC; 7-OH mitragynine; buphedrone; cathinone; 3,4-DMMC; mephedrone; methcathinone; bufotenine; methylone; penytone; ethylone; butylone; naphyrone; pyrovalerone; MDPV; 2C-T-7; 2C-T-2; LSD; TFMPP; atropine; and Bromo-dragonfly. Analytes were considered stable in the matrix under given storage conditions if two of three replicates were positive at a given time point. All samples were analyzed using a Liquid Chromatograph-Time Of Flight (LC/TOF) mass analyzer (Agilent® 1200 HPLC system Agilent TOF 6230 with Jet Stream Technology). The criterion for positivity was acceptable retention time, accurate mass, signal-to-noise ratio, and abundance within 20% of the hand-spiked calibrator.

Drugs were, in general, most stable in urine, with only psilocin, 3-FMC, and 4-FMC failing stability even at room temperature by day 7. Drugs were less stable in blood collected in

gray top tubes maintained refrigerated or frozen. Exceptions to this were cocaine and 2C-H, which failed under refrigerated conditions after two days. Other compounds failed stability in blood at room temperature after two days including 3-FMC, 4-FMC, and psilocin. 4-MEC, buphedrone, cathinone, 3,4-DMMC, mephedrone, and methcathinone failed at seven days at room temperature in blood. Drugs were least stable in serum/plasma, with 3-FMC, flephedrone, naphyrone, and pyrovalerone all failing under refrigerated conditions on day 1, with additional compounds 7-OH mitragynine, 4-MEC, MDPV, mephedrone, and methcathinone also failing on day 7 under refrigerated conditions.

Establishing the stability of drugs intended for inclusion in new analytical procedures is critical as part of validation as many newly emerging drugs are now known to be unstable in certain specimen types and storage conditions after only one to two days. Without information about stability, efforts to interpret toxicological findings can be compromised.

Stability, Stimulant Compounds, Hallucinogenic Compounds

K37 Performance Characteristics of the Neogen ELISA Screening Assay for the Detection of Synthetic Cannabinoids in Urine

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After attending this presentation, attendees will become familiar with the validation and performance characteristics of an Enzyme Linked Immunosorbent Assay (ELISA) urine screening procedure for detection of synthetic cannabinoids in urine.

This presentation will impact the forensic science community by improving interpretation of synthetic cannabinoid immunoassay results and guiding selection of appropriate cutoff concentrations.

Cannabis is the oldest drug of abuse in the world. Synthetic cannabinoids are touted as a legal and safer alternative to cannabis as routine urine drug screening methods do not detect synthetic cannabinoids, making them attractive to users who must undergo random or regular urine drug screenings. The recent emergence of these new legal variants on the market and their widespread availability make it difficult for laboratories to identify and regulatory agencies to prohibit. The goal of this research is to demonstrate that the Neogen® JWH-018 (SPICE) ELISA kit is a highly sensitive and selective method for the rapid detection of JWH-018 N-pentanoic acid in urine.

The Neogen® (SPICE) ELISA kit contained all components required for routine analysis and the assay was performed without modifications according to manufacturer's instruction. Automated analysis was performed on a Freedom EVO® 100 platform configured with a microtiter plate washer and reader.

Performance was evaluated by analyzing results from 2,469 authentic urine samples. Two cutoff concentrations (5 and 10µg/L) were evaluated to classify the samples as positive or negative and establish diagnostic efficiencies. Performance challenges at ±25 and ±50% of these cutoff levels also were investigated to determine intra- and inter-plate imprecision. All 2,469 urine samples were assayed by a qualitative LC/MS/MS for 29 synthetic cannabinoids. Immunoassay results were compared to LC/MS/MS results to determine true positive (TP, positive in both assays for any synthetic cannabinoid), true negative (TN, negative

in both assays), false positive (FP, positive in the Neogen® assay but negative for all synthetic cannabinoids by LC/MS/MS), and false negative (FN, negative in the Neogen® assay but positive by LC/MS/MS). Sensitivity was determined as $(TP/TP + FN) \times 100$, specificity as $(TN/TN + FP) \times 100$, and efficiency as $((TP + TN)/(TP + TN + FP + FN)) \times 100$.

The Neogen® assay was linear from 1-250µg/L with a calculated limit of detection of 0.53µg/L. Intra-plate imprecision (N=7) was <4%, while inter-plate imprecision (N=34) was <9%. Sensitivity, specificity, and efficiency results with the 5µg/L cutoff were 79.9%, 99.7%, and 97.4% and with the 10µg/L cutoff were 69.3%, 99.8%, and 96.3%, respectively. Eighteen of 73 individual, synthetic cannabinoids (25%) exhibited moderate to high cross-reactivity to the target compound. No interferences were present from 94 common drugs of abuse, metabolites, co-administered drugs, over-the-counter medications, or structurally similar compounds.

The absence of extensive sample preparation requirements and short incubation times prove that analysis using the Neogen® (SPICE) ELISA kit, with a high-speed automated system, is a viable method for screening synthetic cannabinoids in urine targeting JWH-018 N-pentanoic acid.

Supported by the Intramural Research Program, National Institute on Drug Abuse, NIH.

Synthetic Cannabinoids, ELISA, Legal Highs

K38 Examination of Toxicology Results in Homicide Victims in New Mexico: 2006-2011

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After attending this presentation, attendees will understand the drug use patterns among homicide victims in New Mexico, as well as the contrast between drugs detected in overdose deaths and those present in homicide victims.

This presentation will impact the forensic science community by exploring the association between alcohol and stimulant drug use and homicidal deaths.

Drug and alcohol abuse are epidemic in the United States. Reviews of the drugs detected in cases of accidental drug toxicity deaths in New Mexico demonstrate very high rates of opiates (heroin and prescription narcotics) and alcohol use leading to death. This research hypothesized that while psychostimulant drugs are only infrequently detected in accidental drug toxicity deaths, the toxicology findings of homicide victims would reflect a different pattern of drug use in the state.

A six-year (2006-2011) retrospective review of toxicology results in homicide victims in New Mexico was undertaken. Exclusion criteria were: (1) age less than 5 years; (2) in-custody deaths; (3) skeletonized remains; (4) no tissue or blood available for toxicology testing; (5) cases where neglect was implicated in causing or contributing to death; and, (6) death from remote injuries. Toxicology results were reviewed and the presence of ethanol, opiates, methamphetamines, cocaine, cannabinoids, and "other drugs" was recorded. Demographic data including age, sex, race, and county of death were recorded. Data was evaluated with SAS[®] statistical software.

During the study period, 983 deaths were certified as homicides. Of those, 67 were excluded by criteria. Of the remaining 916, 18.5% were female and 81.5% were male. Four were 6-11 years old, 47 were 12-17 years old, and 865 were older than 18 years. Neither drugs nor alcohol were detected in the 6-11 years age group. In the 12-17 years age group, there were 38 positive toxicology results (20 for ethanol, 11 for cannabinoids, 4 for cocaine, 2 for "other," 1 for opiates, and 0 for methamphetamine). In the adult (18 years and older) group, there were 1,014 positive toxicology results (488 for ethanol, 134 for cocaine, 126 for cannabinoids, 106 for methamphetamine, 100 for "other," and 60 for opiates). Of adult homicide victims, approximately 12.3% had methamphetamine and 15.5% had cocaine, with 14 cases positive for both methamphetamine and cocaine.

Drugs and alcohol were commonly identified on toxicology testing of homicide victims. Several studies have established the strong association between psychoactive drugs and homicide. These studies have also demonstrated ethanol as the most common substance detected in homicide victims. In this study, ethanol was detected in over half of the cases and opiates were only infrequently detected, which is in sharp contrast to the accidental drug-overdose deaths investigated in New Mexico where opiates were detected most often. Following ethanol, the most commonly detected drugs in homicide victims in the state were stimulants (methamphetamine and cocaine) and cannabinoids. This study's findings support its hypothesis and contribute to the body of literature demonstrating the association between drug use and homicide, particularly among psychostimulant drug users.

Homicide, Toxicology, Stimulants

K39 Postmortem Distribution of 25I-NBOMe After Acute Fatal Toxicity

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After attending this presentation, attendees will learn about the postmortem tissue distribution of 25I-NBOMe in two fatal cases of acute toxicity. The research compound 25I-NBOMe, also known as Cimbi-5 or INBMeO, was developed in academic laboratories as a potent serotonin 2A receptor agonist. Because of its high affinity and ambiguous legal status, recreational drug enthusiasts have sought this compound as a powerful alternative to other hallucinogenic drugs such as lysergic acid diethylamide, or LSD.

This presentation impacts the forensic science community by showing the first forensic characterization of fatalities attributed solely to the toxic effects of 25I-NBOMe. Comprehensive toxicology screens in both cases returned evidence of marijuana use only. A deeper analysis using time-of-flight mass spectrometry revealed the presence of 25I-NBOMe, which was further confirmed by tandem mass spectrometry. The behavior and injuries in these cases reveal a consistent pattern preceding fatal 25I-NBOMe toxicity.

This presentation reports on two deaths following 25I-NBOMe ingestion by decedents who attended separate "rave" parties. Case 1 involved a 21-year-old male who had admitted taking "acid" to his friend. A sudden violent rage caused him to flail about, and subsequently became unresponsive. The postmortem examination revealed numerous external injuries that were consistent with physical aggression. Case 2 involved a 15-year-old female who was socializing outside a rave party, became ill, and rapidly deteriorated as her friend transported her to the hospital. The postmortem assessment was similar to the first case, in which

external contusions were featured prominently. Yet, no anatomical cause of death was found in the internal examination.

Measurements of 25I-NBOMe were accomplished by Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS). In this assay, a liquid-liquid extraction consisting of ethyl acetate was performed on whole blood, stomach contents, liver, bile, brain, vitreous humor, and urine specimens in comparison to 25I-NBOMe-D3 as a deuterated internal standard. After vortexing and centrifugation, the organic layer was collected and acidified, then evaporated with compressed nitrogen until a dry residue remained. The extract was resuspended in mobile phase buffer suitable for analytical injection. The results are shown in the provided table.

Specimen	Case 1 25I-NBOMe (ng/mL)	Case 2 25I-NBOMe (ng/mL)
Whole blood	4.7	16
Vitreous humor	0.0	0.4
Stomach contents	6.5	570
Urine	6.7	19
Liver	0.0	31
Brain	22	241
Bile	17	157

Vitreous humor returned the lowest levels, likely due to lack of distribution during rapid toxicity and a large volume of distribution for 25I-NBOMe. Whole blood, stomach contents, and urine gave acceptable responses, whereas brain and bile were superior specimens, presumably because of their inherently lipid-rich compositions. Because of this property, it is worth noting that brain would seemingly be the best choice for postmortem toxicology analysis, especially under the circumstances of a delayed hospital death.

Without well-characterized scenes and events, these injuries could be perceived as consistent with physical assault resulting in homicide. However, in both cases the internal examination revealed superficial, nonfatal injuries. These findings in the context of the scene characterizations, external body examinations, forensic autopsies, and comprehensive toxicology analyses underscore the necessity of a thorough investigation. Among other 5-HT_{2A} receptor agonists, 25I-NBOMe is not necessarily the most potent since other iterations have been refined through computer simulations and binding activation experiments. New derivatives have been synthesized by replacing the iodine moiety or by substituting the methoxybenzyl ligand on the opposite end of the molecule. Considering these trends, this study anticipates the emergence of many new synthetic derivatives in the near future. For those investigating deaths involving the use of recreational designer drugs, it will be necessary to maintain a level of suspicion that rivals the creativity of those who fabricate these exotic drugs.

2C-I-NBOMe, Designer Drug, Postmortem Distribution

K40 In Vitro Free and Glucuronidated Cannabinoid Stability in Authentic Urine Following Controlled Smoked Cannabis

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After attending this presentation, attendees will be able to describe the phase I and II urinary cannabinoid stability in authentic urine stored for up to one year after controlled cannabis smoking under multiple storage conditions.

This presentation will impact the forensic science community by providing the most extensive cannabinoid stability data to date in authentic urine and aiding forensic toxicologists in cannabinoid test interpretation.

Analyte stability is an important factor in test interpretation. Phase I and II metabolite stability data in authentic urine specimens are limited. Only two studies documented 11-nor-9-carboxy- Δ^9 -tetrahydrocannabinol (THCCOOH) and THCCOOH-glucuronide stability in authentic urine. No studies evaluated Δ^9 -tetrahydrocannabinol glucuronide (THC-glucuronide) urinary stability. Phase I and II cannabinoid stability in authentic urine was evaluated for up to one year after controlled cannabis smoking under multiple storage conditions. This study hypothesized that cannabinoid stability would be affected in a time- and temperature-dependent manner.

Sixteen cannabis smokers (12M, 4F) provided written informed consent to participate in this Institutional Review Board- (IRB) approved study. For each participant, urine specimens collected between 0 and 6h after *ad libitum* cannabis smoking were combined in equal portions to form the high pool; low pool consisted of high pool diluted 1:5 (v/v) with drug-free urine. Pools were stored in polypropylene cryotubes in the dark and baseline concentrations were established by Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS) in triplicate within 24h at room temperature (RT), 4°C and -20°C. Stability was evaluated in duplicate samples after 1 week at RT; 1, 2, 4, 12, and 26 weeks at 4°C and -20°C; and 1 year at -20°C. Limits Of Quantification (LOQ) were: 2.0µg/L for THC and cannabinol; 1.0µg/L for THCCOOH, 11-hydroxy-THC (11-OH-THC), and cannabidiol; 0.5 µg/L for THC-glucuronide; and 5.0µg/L for THCCOOH-glucuronide. Stabilities at different storage times and temperatures compared to those at baseline were evaluated with Wilcoxon matched-pairs signed-ranks test (RT) or repeated-measures Friedman test (4°C and -20°C); Dunn's multiple comparisons tests were used for *post hoc* comparisons. Results with 2-tailed P<0.05 were considered significant.

At baseline, THCCOOH, THC-glucuronide, and THCCOOH-glucuronide were quantified in: 12, 16, and 15 low RT pools; 13, 16, and 15 high RT pools; 7, 16, and 15 low 4°C pools; 12, 16, and 15 high 4°C pools; 5, 15, and 15 low -20°C pools; and 12, 16, and 15 high -20°C pools, respectively. THC, cannabidiol, and cannabinol were never detected. RT THCCOOH baseline concentrations were significantly higher than at -20°C, but not 4°C. After one week at RT, THCCOOH increased, THCCOOH-glucuronide decreased, but THC-glucuronide remained unchanged. In RT low pool only, total THCCOOH (molar sum of THCCOOH and THCCOOH-glucuronide) was significantly lower after one week. At 4°C, THCCOOH was stable up to two weeks, THCCOOH-glucuronide up to one month, and THC-glucuronide for at least six months. At -20°C in both pools, THCCOOH was stable at one year, but high pool results at six months were high; THC-glucuronide and THCCOOH-glucuronide were stable for six months. Total THCCOOH was stable for at least six months at 4°C, and up to six months (low) and at least one year (high) at -20°C. Although not detected initially, 11-OH-THC was detected in two low and three high pools after one week at RT. These are the most extensive cannabinoid stability data to date in authentic urine.

Substantial THCCOOH-glucuronide deconjugation was observed at RT and 4°C and frozen specimen storage provided

maximum stability, with cannabinoid glucuronides and total THCCOOH being stable for six months.

Supported by the National Institutes of Health, IRP, National Institute on Drug Abuse.

Cannabinoids, Glucuronides, Stability

K41 Uncertainty of Blood Alcohol Concentration (BAC) Results as Related to Instrumental Conditions: Optimization and Robustness of BAC Analysis Parameters

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After attending this presentation, attendees will learn the concepts and practices of the quality-by-design approach to analytical method development to ensure both optimal and robust method conditions. Specifically applied to the analysis of Blood Alcohol Concentration (BAC), this approach will be used to determine optimal chromatographic and headspace conditions to yield a robust set of method parameters with determination as to how these parameters affect both detection limits and uncertainty. Finally, the presentation will extend these concepts to the two most common types of headspace samplers used for BAC analysis: pressurized loop systems and volumetric systems.

This presentation will impact the forensic science community by providing a better understanding of how the major instrument parameters affect both detection limits and uncertainty of headspace gas chromatographic analyses, specifically for BAC analysis. This approach can, and should, be used for many applications in analytical laboratories to ensure that the instruments are operated in a manner that allows for normal variations to occur without an impact to the final determinations. In short, if proper robustness experiments have not been conducted, it is possible the normal daily variance in instrument parameters could cause non-compliance or a lack of determinative value control, which may not be measured by normal system Quality Control (QC) samples. For this reason, robustness experiments should be common in the initial method validation of analytical measurements.

Analysis of blood alcohol concentration is a routine analysis performed in many forensic laboratories. This analysis commonly utilizes headspace-sampling, followed by Gas Chromatography combined with Flame Ionization Detection (GC/FID). Studies have shown several "ideal" methods for instrumental operating conditions which are intended to yield accurate and precise data. Given that different instruments, sampling methods, application-specific columns, and parameters are often utilized, it is less common to find information on the robustness of these reported conditions. A major problem can arise when these "ideal" conditions may not also be robust, thus producing data with higher-than-desired uncertainty or inaccurate results.

The goal of this research is to incorporate the principles of Quality By Design (QBD) in the development of BAC instrument parameters, thereby ensuring that minor instrumental variations, which occur as a matter of normal work, do not appreciably affect the final results of this analysis. This presentation will discuss both the QBD principles as well as the results of the experiments which allow for determination of "ideal" instrumental conditions. Additionally, method detection limits will also be reported in order to determine a reporting threshold and the degree of uncertainty at the common threshold value of 0.08g/dL. Finally, differences between pressurized loop headspace systems and volumetric headspace systems will be discussed, comparing and contrasting

these two different types of analytical instruments.

Blood Alcohol Concentration, Robustness, Optimization

K42 Influence of the Genetic Polymorphism in CYP2C9 on the Pharmacokinetics of Delta-9-Tetrahydrocannabinol and Introduction of a New Pharmacokinetic Model

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After attending this presentation, attendees will understand that pharmacokinetic differences in blood concentrations of cannabis smokers depend heavily on the genetic polymorphism in the hepatic enzyme CYP2C9. Additionally, based on a new non-linear mixed-effects pharmacokinetic model developed, a simple yet highly accurate predictive model will be introduced at the presentation which allows for the estimate of time of consumption after a single consumption using Excel®.

This presentation will impact the forensic science community by stimulating the interest of forensic toxicologists to the field of pharmacogenomics and its importance to the interpretation of the analytical results (e.g., for Driving Under the Influence (DUI) cases).

Background: Medical and recreational use of *Cannabis sativa* is on the rise worldwide. There is a need for a better understanding of inter-individual differences in the pharmacokinetics of cannabinoids. Degradation of orally ingested Δ^9 -tetrahydrocannabinol (THC) to the pharmacologically active metabolite 11-hydroxy-THC (11-OH-THC) and 11-nor-9-carboxy-THC (COOH-THC) largely depends on hepatic P450 activity.¹ In this study, the impact of the CYP2C9 polymorphism on the pharmacokinetics of intravenously administered Δ^9 -tetrahydrocannabinol was studied in healthy volunteers who were long-time abstainers from cannabis or were cannabis naive.

Methods: Three hundred and six healthy volunteers were screened for CYP2C9 polymorphisms and phenotyped for CYP2C9 single nucleotide polymorphisms *2 (Arg144Cys) and *3 (Ile359Leu).

According to genetic subgroups, this study included 25 volunteers (11 males, 14 females). Heparinized blood samples were drawn arterially before and in short intervals up to five hours after a single intravenous bolus of 0.1mg/kg THC. Additional samples were taken from venous blood 24 and 48 hours afterward.

Sample preparation involved a liquid/liquid extraction (ethyl acetate:cyclohexane = 1:7). THC, 11-OH-THC, and COOH-THC were quantified by Liquid Chromatography with Tandem Mass Spectrometry (LC/MS/MS). The Limits of Detection (LODs) were 0.08ng/mL for THC, 0.18ng/mL for 11-OH-THC, and 1.4ng/mL for COOH-THC. The Limits of Quantification (LOQs) were 0.76ng/mL for THC, 0.48ng/mL for 11-OH-THC, and 2.0ng/mL for COOH-THC. Linearity for the 8-point calibrations extended to 600ng/mL for THC and for COOH-THC, and to 60ng/mL for 11-OH-THC. For the quantification, deuterated internal standards were used for all analytes.

Data were analyzed using Phoenix® NLME™ V1.3 and NCSS™ 8.0.14.

Results and Conclusion: Five out of eight possible different mutations were found in the 306 volunteers analyzed. Fifty-nine percent were wildtype CYP2C9*2 and *3. Δ9-THC half-times were dependent on genotypes and varied by two hours during the terminal elimination phase. There is a slow metabolizer phenotype for CYP2C9*3.

The COOH-THC metabolite accumulates to exceed the concentrations of the parent by up to 100-fold in individuals with wild type CYP2C9. These findings show there is a slow metabolizer phenotype for CYP2C9*3, which changes the metabolite ratio of THC:COOH-THC from 10:1 to 2:1. These results have consequences for the use of metabolite ratio predictive methods in forensic casework.

Genetic variability determines the pharmacokinetics of THC with impact on vital signs and psychotropic side effects. These findings will allow a more reliable interpretation of cannabinoid blood concentrations in forensic and clinical cases.

Reference:

1. Sachse-Seeboth C, Pfeil J, Sehr D et al. Interindividual variation in the pharmacokinetics of delta-9-tetrahydrocannabinol as related to genetic polymorphisms in CYP2C9. *Clin Pharmacol Ther* 2009;85:273-6.

Tetrahydrocannabinol, Pharmacokinetics, Pharmacogenomics

K43 Utilizing Protein Adducts as Exposure Markers for Nitrogen Mustard Chemical Warfare Agents

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After attending this presentation, attendees will have a better understanding of protein adducts and how these adducts can be used as exposure markers for chemical warfare agents.

This presentation will impact the forensic science community by demonstrating the advantages of protein adduct biomarkers as compared to more traditional methods of exposure determination.

This presentation investigates protein adduction by nitrogen mustard chemical warfare agents in the initial development of longer-term markers of human exposure to these compounds. Protein adducts are formed when electrophilic xenobiotics (either a parent compound or a metabolite) bind to nucleophilic amino acids in biological proteins. The three most important protein nucleophiles for xenobiotic adduction are thiol group of cysteine (Cys) and the amino groups of lysine (Lys) and histidine (His). The selectivity of adduction to these sites is in part dependent on the physicochemical nature of the electrophile in question.

The chemical warfare agents mechloroethamine (HN-2) and tris-(2-chlorethyl)amine (HN-3) are nitrogen mustards that can cause tissue blistering and ocular/respiratory damage following human inhalation and dermal exposure. Previous *in vitro* work in this study's laboratory demonstrated the ability of HN-2 and HN-3 to bind to Cys, Lys, and His residues in model peptides, in addition to His and Lys residues on purified hemoglobin (Hb) and Human Serum Albumin (HSA). The objective of this current work was to confirm and identify adducts on Hb and HSA after *in vitro* incubation with HN-2 and HN-3 in whole blood.

Adduct formation was induced by incubating human whole blood aliquots for 24h at 37°C with HN-2 and HN-3 at a molar excess as compared to protein nucleophile. Hb and HSA were extracted from whole blood via an initial centrifugation to separate

erythrocytes (containing Hb) from plasma (containing HSA). Proteins were then extracted via precipitation with acidic acetone (Hb) or acidic alcohol (HSA). Extracted proteins were quantified using the Bradford protein assay, digested using trypsin, and the resulting tryptic peptides were separated and analyzed using Ultra High Performance Liquid Chromatography-Tandem Mass Spectrometry (UHPLC-MS/MS). Analysis was performed on an Agilent® 6530 Quadrupole Time-Of-Flight (QTOF) MS equipped with a 1290 Infinity® UHPLC and a ZORBAX Eclipse Plus C-18 Rapid Res HD (10mm x 2.1mm, 1.8µm particle size) column. Gradient elution of tryptic peptides with water and acetonitrile mobile phases containing 0.1% trifluoroacetic acid (TFA) was utilized. Agilent® Mass Hunter BioConfirm Software was used to identify specific sites of adduction. HN-2 and HN-3 adduction was observed on Hb and HSA, similar to what was seen in previous work using purified proteins incubated with these agents. Identified adducts were hydrolysis products of HN-2 and HN-3, most commonly found on His residues residing on the surface of both HSA and Hb. Adduction was confirmed using replicate samples, exact mass MS, and MS/MS analysis. Identified adducts were determined to be stable at 37°C for up to one week after incubation with HN-2 and HN-3.

The use of protein adducts can, in theory, allow for detection of HN-2 or HN-3 exposure long after urinary metabolites and/or parent compounds have been excreted from the body. This is of significance to the forensic community, as current methods focus on blood or urinary metabolites for exposure determination, which may suffer from certain drawbacks such as non-specificity and short detection window based on the half-life of the compounds of interest. It is of high importance to be able to identify previous exposure to these dangerous compounds, in terms of both victim exposure and in those individuals who may have been involved in unlawful handling or synthesis of these compounds.

Chemical Warfare Agents, Exposure Biomarkers, Nitrogen Mustards

K44 Plasma and Oral Fluid l-Methamphetamine Concentrations After Controlled Vicks VapoInhaler® Administration

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After attending this presentation, attendees will be able to describe l-methamphetamine (l-MAMP) pharmacokinetics in plasma and oral fluid (OF) after intranasal administration according to manufacturer's recommendations of over-the-counter Vicks® VapoInhaler™.

These findings will impact the forensic science community by improving interpretation of MAMP and amphetamine (AMP) OF results, guiding selection of MAMP OF screening tests, and performing chiral separation of MAMP and AMP for confirmation when needed.

AMPs are widely abused psychoactive substances included in drug testing programs in forensic, workplace, drug abuse treatment, and anti-doping settings. Consumption of over-the-counter Vicks® VapoInhaler™ can produce positive blood and urine MAMP results, requiring chiral separation of d- and l-enantiomers in positive cases. l-MAMP is the primary active component in Vicks,

with the manufacturer reporting potential trace *d*-MAMP presence. To date, there are no published OF MAMP or AMP data following controlled Vicks® administration.

Thirteen healthy adults (ten male, three female) aged 19-54 years old participated in this National Institute on Drug Abuse Institutional Review Board-approved study; all provided written informed consent. Participants arrived the morning of the first dosing day (Day 1), when two sprays per nostril every 2h (manufacturer's dosing recommendation) were administered from 9:00 a.m. until 7:00 p.m. (six doses). A single dose was administered at 6:00 a.m. on Day 2. The manufacturer suggests that 0.16-0.6mg *l*-MAMP is administered with each dose. Plasma and OF specimens were collected before and up to 32h after the first dose throughout the two days. Plasma was collected with an indwelling venous catheter, and OF was collected with the Oral-Eze® collection device and was also collected for the Dräger DrugTest® 5000 on-site drug test. *d*- and *l*-MAMP and AMP enantiomers were confirmed in plasma and Oral-Eze® specimens by a Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS) method after solid phase extraction and derivatization with 1-fluoro-2,4-dinitrophenyl-5-*l*-alanineamide (Marfey's reagent). The method's Limit Of Quantitation (LOQ) for all analytes in both matrices was 1µg/L.

A total of 351 plasma specimens from 13 participants and 324 OF specimens from 12 participants were collected. *d*-MAMP and *d*-AMP were not detected in any plasma or OF specimen. Only 2 participants were positive for *l*-MAMP in plasma, producing a combined 52 positive specimens at the method LOQ (14.8%). Maximum *l*-MAMP plasma concentrations did not exceed 10µg/L, and both participants were still positive 11h after the final dose. No *l*-AMP was present in plasma. The total number of OF specimens positive for *l*-MAMP at the method LOQ (1µg/L), and the 2004 proposed Substance Abuse and Mental Health Services Administration (SAMHSA) screening (AMPs 50µg/L) and confirmation (MAMP 50µg/L and AMP >LOD) cutoffs were 262 (80.9%, N=12 participants), 24 (7.4%, N=2), and 15 (4.6%, N=1), respectively. Suggestions were made to lower the screening and confirmation cutoffs to 25µg/L, similar to the Driving Under the Influence of Drugs, Alcohol, and Medicines (DRUID) cutoffs. At these lower 25µg/L cutoffs, there were 31 positive OF specimens (9.6%, N=3). The median (range) maximum *l*-MAMP concentration was 16.7 (8.4-182) µg/L. One participant had detectable OF *l*-AMP at the method LOQ, first detected 7h after the first dose. The maximum *l*-AMP concentration observed was 5.5µg/L. At the method LOQ, 7 participants (58.3% specimens) were still positive for *l*-MAMP 11h after the last dose; among the other 5, none were positive 9h after the last dose. At the proposed SAMHSA screening cutoff of 50µg/L, one participant was only *l*-MAMP positive in one sample immediately after the second dose (N=1), and another participant remained positive at the last OF specimen 11h after the last dose. At the confirmation cutoff, the last detection time was >11h after the last dose (N=1). The DrugTest 5000® utilizes a 35µg/L *d*-MAMP cutoff; all participants' OF specimens were negative on this test.

These data suggest that careful selection of an AMP's screening test for OF targeted toward *d*-MAMP may obviate the need for chiral separation and confirmation of presumptive positive AMPs tests. Furthermore, these data provide an LC-MS/MS confirmation method for *d*- and *l*-AMPs in OF that utilizes a chiral derivative rather than a chiral column. These data provide valuable new data for interpretation of OF AMPs tests.

Supported by the Intramural Research Program, National Institute on Drug Abuse, NIH, and the Substance Abuse Mental Health Services Administration (SAMHSA).

L-Methamphetamine, Oral Fluid, Chiral Separation

K45 Evaluation of On-Site Oral Fluid Drug Screening Using the Dräger Drug Test® 5000 and Affiniton® Drugwipe® in Suspected Impaired Drivers

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After attending this presentation, attendees will be able to assess the utility of on-site oral fluid drug testing devices designed for use in the field and their ability to generate results that can later be confirmed in the laboratory in a preserved oral fluid specimen or a more traditional specimen such as blood or urine.

This presentation will impact the forensic science community by providing data on the utility and reliability of point-of-contact oral fluid drug tests in suspected impaired drivers. By comparing the presumptive roadside results to laboratory confirmation results, which is critical to ensure their admissibility in court, the overall accuracy of these field tests were determined.

The purpose of this project was to evaluate on-site oral fluid drug testing devices and compare the presumptive results to laboratory-based oral fluid confirmatory results as well as to blood or urine results in certain cases.

The ease of obtaining an oral fluid sample in the field proximate to the time of driving has made it an ideal specimen for roadside testing in drug-impaired driving investigations. Two point-of-contact oral fluid testing devices, the Dräger Drug Test® 5000 (DDT 5000) and/or the Affiniton® Drugwipe® (Drugwipe), were evaluated in two separate roadside studies. Subjects were recruited into the study after the conclusion of their arrest for suspected impaired driving. The officers followed their routine arrest procedures, including advisement of rights, field sobriety tests, portable breath test, blood sample or urine collection, and completion of the arrest paperwork before offering the subjects the opportunity to provide oral fluid samples for research purposes. Samples were collected for the DDT 5000 and/or Drugwipe devices following their individual manufacturer-recommended protocols. Following provision of the DDT 5000 and Drugwipe samples, participants were asked to provide an additional oral fluid sample collected with an Immunalysis Quantisal™ collection device for laboratory-based confirmatory analysis. Both devices included tests for amphetamine, methamphetamine, cocaine, opiates, benzodiazepines, and THC. The DDT 5000 device also tested for methadone.

All oral fluid samples were analyzed to confirm the presence of target drugs routinely screened for in drug-impaired driving cases. The laboratory-based test was considered the "true result," and the field test results for that subject were evaluated against that with each given matrix. The overall effectiveness of the field test was based on sensitivity, specificity, and accuracy for both the DDT 5000 and Drugwipe, relative to the laboratory-based confirmation result.

At one study location, the DDT 5000 was compared to laboratory-based oral fluid confirmations in a total of 79 cases. The overall sensitivity, specificity, and accuracy were 50.0%, 99.8%, and 94.8%, respectively. The overall sensitivity, specificity, and accuracy of the Drugwipe were 48.9%, 100%, and 93.8%, respectively. In comparison, the overall sensitivity, specificity, and accuracy of the DDT 5000 in 33 cases compared to confirmed analytes in oral fluid were 61.4%, 99.2%, and 87.7%, respectively at the other location. Additionally, the overall sensitivity, specificity, and accuracy of

the DDT 5000 compared to compounds confirmed in blood were 72.2%, 98.6%, and 93.5%, respectively. In comparing the 16 cases where both blood and oral fluid were analyzed and using blood as the "true result," overall sensitivity, specificity, and accuracy were 78.9%, 98.6%, and 94.6%, indicating good correlation between analytes confirmed in oral fluid to those confirmed in blood. For some drugs, prevalence was very low (e.g., amphetamines and methamphetamine), preventing meaningful calculations of sensitivity and specificity.

The use of oral fluid drug-testing devices, like the DDT 5000 and Drugwipe, offer the ability to rapidly obtain a screening result in the field at the time of a stop, which may provide useful data that can aid in the investigation of a drug-impaired driving case. Although the DDT 5000 and the Drugwipe devices had different cut-offs for individual drug classes within their scope, they performed comparably in terms of overall sensitivity, specificity, and accuracy in actual drug-using subjects. Although sensitivity will have to be improved for comprehensive detection of drug use in drivers, the current generations of devices are good tools for deterrence and minimizing the risk of false positive results.

Oral Fluid, On-Site Testing, Drug-Impaired Driving

K46 Detection of Carboxylated Metabolites of XLR-11, UR-144, and Their Pyrolysis Products in Oral Fluid

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The goal of this presentation is to inform attendees about the complexity of analyzing thermally unstable synthetic cannabinoids like XLR-11 and UR-144. The possibility of monitoring synthetic cannabinoid metabolites in oral fluid and its potential applications will be discussed.

This presentation will impact the forensic science community by demonstrating that synthetic cannabinoid metabolites can be detected in oral fluid specimens and their detection can possibly prove useful in avoiding passive exposure defense in drug court cases.

Introduction and Objectives: Synthetic Cannabinoids (SC) are the most frequently found group of emerging drugs in routine testing. Like marijuana, they are ingested by smoking, produce similar subjective effects, and, hence, are referred to as "Synthetic Marijuana." XLR-11 and UR-144 are currently the most popular but thermally unstable components in "Synthetic Marijuana" preparations. In this study, detection of carboxylated metabolites of both UR-144 and its pyrolytic product (UR-144 3,3,4-trimethyl pentenoyl isomer) are reported for the first time. Seventy-four oral fluid specimens previously confirmed positive (32) or negative (42) by Liquid Chromatography with Tandem Mass Spectrometry (LC/MS/MS) for parent drugs XLR-11 and/or UR-144 were re-analyzed for the presence of UR-144 N-pentanoic acid and UR-144 degradant pentanoic acid, common metabolites of XLR-11 and UR-144.

Methods: Oral fluids were collected using the Quantisal™ device, resulting in a 1:4 dilution. Standard reference materials for carboxy metabolites of UR-144 and its pyrolytic product were obtained from Cayman Chemical®. Acid metabolites for the UR-144 and its pyrolysis product were observed when human liver microsomes were incubated with commercially available XLR-11 and XLR-11 degradant respectively for a separate study,

demonstrating these to be common metabolites for XLR-11 and UR-144. Carboxylated metabolites were extracted from 0.5mL of acidified oral fluids (60µL phosphate buffer added, pH 1.8) by liquid-liquid extraction with hexane:ethyl acetate (7:1). The top layer was dried down and re-constituted with 50µl methanol for a 20µL injection onto the LC/MS/MS system in negative Multiple Reaction Monitoring (MRM) mode using Electrospray Ionization (ESI). Separation was performed on a 5µ biphenyl column with 0.7mL/minute flow rate. Mobile phases were 0.1% formic acid with 2mM ammonium formate and 0.1% formic acid with 2mM ammonium formate in acetonitrile. The gradient started at 20% organic, was held for 0.5 minutes and increased to 50%, 60%, and 90% after 0.6, 2.9, and 3 minutes, respectively, before returning to initial conditions at 4 minutes. The method was applied to 74 authentic oral fluid specimens with an administrative cut-off of 10pg/mL.

Results: Full-scan positive mode ionization MS data from microsome incubations of XLR-11 degradant showed a metabolite with the same mass and similar fragmentation to that of UR-144 N-pentanoic acid. In negative ion MRM mode, the same precursor and product ions as the UR-144 N-pentanoic acid standard were present but eluted earlier than the standard. Therefore, this metabolite was first presumptively identified as carboxylated UR-144 degradant and later confirmed by comparison with the synthesized standard. In oral fluids tested, UR-144 N-pentanoic acid metabolite was detected at or above 10pg/mL in 15 (50%) of the 32 specimens positive for parent and 2 (5%) of the 42 negative specimens. The concentrations of the UR-144 acid metabolite ranged from 15-500pg/mL. The carboxylated metabolite for UR-144 degradant was also detected in all specimens positive for UR-144 N-pentanoic acid.

Conclusions: This is the first study reporting detection of carboxy metabolites of the most prevalent synthetic marijuana ingredients XLR-11 and UR-144. Their quantitative evaluation in oral fluid opens up possibilities for application as markers for SC abuse. Further research on their application and perhaps more sensitive techniques are needed to fully exploit their potential in oral fluid testing. Monitoring oral fluid for SC metabolites could prove useful in minimizing passive exposure defense in drug testing cases.

Synthetic Cannabinoids, XLR-11/UR-144, Pyrolysis

K47 LC/MS/MS Quantification of Ethyl Glucuronide and Ethyl Sulfate in Meconium

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After attending this presentation, attendees will be able to describe sample preparation, Limits Of Quantification (LOQ) and Detection (LOD), extraction efficiency, matrix effect, accuracy, and imprecision of this Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS) Ethyl Glucuronide (EtG) and Ethyl Sulfate (EtS) method.

This presentation will impact the forensic science community by describing an analytical method that offers new approaches to sample clean-up and chromatographic separation of EtG and EtS in meconium. Investigators need alternatives to Fatty Acid Ethyl Esters (FAEE) to identify and quantify *in utero* alcohol exposure; this method for quantifying EtG and EtS in meconium offers a less problematic approach to identify *in utero* alcohol

exposure.

Introduction: Maternal alcohol consumption during pregnancy is associated with fetal alcohol spectrum disorder that encompasses growth retardation, craniofacial dysmorphism, cognitive disorders, and social impairments. According to the 2011 National Surveys of Drug Use and Health report, 9.4% of pregnant respondents 15-44 years old admitted to current alcohol use, with 2.6% reporting binge drinking. A small percentage of ingested ethanol undergoes non-oxidative metabolism yielding FAEEs, EtG, and EtS. These chemical ethanol derivatives are detectable longer than ethanol in several matrices and are, therefore, clinically useful in identifying recent alcohol exposure. Alcohol marker testing in meconium, the first neonatal feces, can identify *in utero* alcohol exposure during the 3rd and perhaps 2nd trimesters. EtG and EtS are thought to prevent false positive reporting of maternal alcohol consumption during pregnancy, whereas FAEE may be present in meconium due to olive oil intake during pregnancy, and are highly unstable in meconium unless specimens are frozen from collection to analysis.¹ Several methods describe meconium FAEE quantification, although currently only one published method reports quantification of EtG and EtS in meconium. This study proposed to develop and validate a selective and sensitive LC/MS/MS assay for EtG and EtS in meconium.

Methods: Blank meconium (0.1 ± 0.003g) was fortified with d5-EtG and d5-EtS internal standards. Specimens were homogenized, vortexed, centrifuged, and supernatant transferred to clean tubes. Meconium supernatant was loaded onto a pre-conditioned Biotage anion-exchange column (100mg/3mL). Columns were washed with acetonitrile and methanol, and analytes eluted with 1% hydrochloric acid in acetonitrile. Extracts were dried under nitrogen at 40°C and reconstituted in water with 0.1% formic acid. An ABSciex 5500 Qtrap® mass spectrometer was interfaced with a Shimadzu UFLCXR system for analysis with gradient chromatographic separation of EtG and EtS on a Phenomenex Kinetex® XB-C18 column (2.1x 100mm, 2.6µm). Mobile phase was water and methanol, modified with 0.1% formic acid. Sensitivity, specificity, linearity, accuracy, imprecision, extraction efficiency, matrix effect, carryover, and dilution integrity were included in the method validation. Meconium samples from infants whose mothers self-reported alcohol consumption during pregnancy were analyzed, demonstrating method applicability.

Results: Linear ranges for EtS and EtG were 2.5-500 and 5-1000ng/g, respectively. Calibration curves employed 1/x² weighting (correlation coefficients ≥0.990). Extraction efficiencies across the linear range were 56.5-84.6% for EtS and 60.6-72.1% for EtG. Matrix effects were -18.6% to 28.9% for EtG and EtS. Analytical accuracy was 101.9-112.6% for both analytes at three quality control (QC) concentrations across the linear range; inter-day imprecision (%CV) was 2.2-11.0% for both analytes (N=18). Meconium from six different negative pools contained no interfering peaks. None of 92 potential exogenous interferences fortified at 10,000ng/g into low Quality Control (QC) samples caused quantification criteria or transition ratios to fail for either analyte. No carryover was detected after a specimen containing two times the upper LOQ. Analytes were stable (≤15% concentration change) on the autosampler at 4°C after 72h.

Conclusions: This analytical method offers new approaches to sample clean-up and chromatographic separation of EtG and EtS in meconium. Investigators need alternatives to FAEE to identify and quantify *in utero* alcohol exposure; this method for quantifying EtG and EtS in meconium offers a less problematic approach to identify *in utero* alcohol exposure.

Supported by the Intramural Research Program, National Institute on Drug Abuse, NIH

Reference:

1. Chan, D, et al. (2003). "Population baseline of meconium

fatty acid ethyl esters among infants of nondrinking women in Jerusalem and Toronto." *Ther Drug Monit* 25(3): 271-278.

Meconium, Ethyl Glucuronide, FAEE

K48 Screening for Drugs and Metabolites in Hair Specimens by LC/TOF/MS

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After attending this presentation, attendees will gain knowledge about the contribution of hair as a matrix for the detection of drugs using Liquid Chromatography Time-of-Flight Mass Spectrometry (LC/TOF/MS) and how these results compare to those previously reported for other postmortem biological matrices originating from the same decedent.

This presentation will impact the forensic science community by establishing a procedure for the screening of drugs and metabolites in hair by LC/TOF/MS and recognizing hair as a valuable alternative matrix for drug screening when blood and urine sample collection is delayed or unavailable within the detectable window for those matrices.

The mechanism by which drugs are incorporated into hair has been most simply modeled as passive diffusion into the rapidly growing cells of the hair follicle; however, alternative studies describe mechanisms for incorporation that occur post-formation of the hair shaft that may be drug-dependent. Taken together, forensic analyses largely benefit from the very stable matrix that the interior of the hair shaft provides, thus allowing for a detection window that is days to years post-exposure. To this end, several studies have described methodologies for the analysis of hair, most recently utilizing the specificity and data range accomplished with time-of-flight mass spectrometry.

Because of advances in technology and affordability, LC/TOF/MS has gained increasing popularity as a screening tool for its ability to determine mass measurements that are accurate to several decimal places (rather than the nominal or unit mass measurements of ±0.01 Da) coupled with the creation of a target drug compound list with retention time data which allows for rapid, selective, specific, and retrograde screening of target compounds. To this end, this study's laboratory has recently validated an LC/TOF/MS screening method and has created an Accurate-Mass Retention-Time (AMRT) Personal Compound Database Library for more than 100 pure reference standard drug compounds routinely and newly encountered in forensic toxicology examinations.

This study describes the procedure developed in the laboratory for the analysis of drugs extracted from hair by LC/TOF/MS. Postmortem head hair samples were collected from a variety of medicolegal cases for which chronic drug use had been documented, and forensic analyses on blood, urine, and other routine toxicology specimens from the same case had been completed by the laboratory. This presentation will highlight five interesting cases involving scene information and toxicology findings in blood, hair, and other specimens. Approximately 10mg from the total hair sample was subjected to a decontamination/wash step with a solution of 0.1% Triton™ X-100, and subsequently extracted with an extraction buffer composed of 200mM dithiothreitol with gentle rocking at room temperature for two hours. The final external wash and internal extracts were combined with internal standard, filtered, and analyzed using LC/TOF/MS to scan for the presence of prescription, illicit, and newer designer drugs.

Separations were performed using an Agilent® Eclipse Plus

C18 1.8um, 3.0 x 100mm column. The chromatography conditions were able to resolve isomers such as codeine and hydrocodone, but could not differentiate between 3,4-methylenedioxy-methamphetamine (MDMA) and methedrone. In the case of overlapping isomers, supportive information was obtained from distinguishing accurate mass measurements of associated metabolites. The mass analyzer was an Agilent® 6230 TOF/MS operated in positive ion scan mode with mass scanning from 100 to 1,000m/z.

This presentation will discuss the significant strengths and weaknesses of this technique in comparison to traditional methods of analysis. However, this type of comprehensive toxicology analysis will extend the abilities of laboratory operations and provide more interpretive information for the forensic toxicologist. The analytes discovered in hair could also reveal a medicinal history that would be useful to a pathologist in classifying death when very little else is known or proven.

Hair Analysis, LC/TOF/MS, Drugs of Abuse

K49 Postmortem Pediatric Toxicology

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After attending this presentation, attendees will gain an appreciation for the challenges unique to toxicological findings in postmortem pediatric cases. Attendees will learn interpretive guidelines for pediatric cases involving forensic toxicology in both a general and case-specific sense.

This presentation will impact the forensic science community by further delineating the interpretive aspects of toxicological findings in the pediatric population.

In this 15th Annual Special Session within the Toxicology section, pediatric cases involving toxicological findings are discussed. As a relative dearth exists of interpretive information involving toxicological findings in the pediatric population, this session is a forum to help elucidate and clarify such issues. The format is a short case presentation including pharmacokinetic data and other relevant ancillary information followed by audience participation to provide interpretive clarity around the case-specific impact of the toxicological findings. This session, attended by various sections of the Academy, allows for various perspectives of case issues that lead to integrative consensus, or differing opinions, as to cause of death in children.

Gregory Davis, MD, will be presenting a case involving the death of a 16-year-old who purchased, via the internet, a substance called "Blue Nitro." The active ingredient in this concoction was gamma-hydroxybutyric acid (GHB). This substance, originally developed as a potential anesthetic agent and whose abuse for everything from weight loss to sedation to androgenic properties, is potentially lethal. It is a challenge analytically as it is not part of most routine toxicological screens. Interpretive issues include consideration of postmortem formation of the substance. Dr. Davis will discuss the role of this substance in the individual's death.

Andrea McCollum, MD, will be speaking to a case of a 7-week-old female who was found expired face down in bed with her mother at home at 11:20 p.m. She had been fed at approximately

4:00 p.m. on February 5, 2013. The previous day, she had been dropped from a waist-level height by her 17-year-old sister. There were no apparent signs of trauma as a result of the fall, according to the mother. Toxicology showed 1.82mg/L diphenhydramine in blood with a qualitative (positive) result for fluconazole. The case will highlight a combination of factors, including the issues of an infant sleeping in the same bed as an adult juxtaposed to the toxicological findings. The use of diphenhydramine in children has been widespread based on its sedative properties and the conception of safety as an over-the-counter substance.

William Anderson, PhD, will discuss a homicidal poisoning case of a child with disulfoton, an organophosphate pesticide. A sibling was also poisoned, but survived. The compound was administered in a flavored drink, thus attempting to hide its potentially offending taste. The case will highlight the clinical, toxicological, analytical, and forensic aspects of this toxicant. It will also stress the need for toxicologists and pathologists to be aware of substances other than drugs.

Sally Aiken, MD, will present a case of an infant that was admitted to a hospital with subdural hemorrhage, thought to be child abuse, but with an unusual presentation. Eventually, the child was put into hospice care, where it was given phenobarbital for sedation. The child died with a very high phenobarbital concentration postmortem.

Pediatric, Postmortem, Toxicology



Last Word Society



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LW1 The Pazzi's Conspiracy: An Investigation of a 500-Year-Old Attempted Coup D'État in Renaissance Florence

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After attending this presentation, attendees will understand how it is possible to investigate ancient murders and conspiracies from a forensic point of view to reconstruct historical facts.

This presentation will impact the forensic science community by suggesting a new and more exhaustive reconstruction of the crime dynamic of an attempted coup d'état against the Medici family and Lorenzo the Magnificent, during which his brother Giuliano was assassinated.

The reasons for the plot can be explained through the context of the political state at the time, reminding us of how the imbalance of power among the Italian city-states began. Two years before the incident, the Medicis' close ally, the Duke of Milan Galeazzo Sforza, had been murdered, leaving his scheming counselor, Cicco Simonetta, to block Florence's future need for defense. With the revelation of the Pazzi conspiracy, Lorenzo was indeed isolated, especially in light of new evidence pointing to the collusion of the Duke of Urbino, Federico da Montefeltro. An encrypted letter written by Federico was unearthed from a private archive in Urbino and decoded with the help of Cicco's Rules for Extracting Ciphred Letters Without a Sample. The letter revealed a ruthless Machiavellian intent to seize Florence. After the plot's failure, Cicco fell to usurpers of the Milanese state and Lorenzo had to scramble for support by groveling to the kingdom of Naples and patching things up with Rome; Lorenzo would later have his revenge, through Botticelli's allusion to the murder and conspiracy in works such as Primavera.

In the Cathedral of Florence during Ascension Sunday Mass in 1478, the two powerful leaders of the Florentine state, Lorenzo and Giuliano de' Medici, were attacked. Stabbed 19 times, Giuliano died; Lorenzo was wounded, but he survived. Members of the rival Pazzi family and their supporters were subsequently captured and variously disposed of.

To reconstruct the conspiracy events and Giuliano's murder, the crime's descriptions by the eyewitnesses, preserved in ancient publications needed to be compared, with physical pieces of evidence from the victim's body. Giuliano's remains had been exhumed in 1947 by the anthropologist and physician Giuseppe Genna and the physician Gaetano Pieraccini, but, unfortunately, their scientific reports were only partially published and the whole documentation seemed lost after their deaths. A long biographic research, following the clues provided by Genna's private story, allowed the recovery of his archives in Rome: pictures, X-rays, handwritten notes, and plaster casts could be eventually examined under the modern forensic magnifier.

The wounds shown in the pictures and the cast of Giuliano's skull bear evidence of plastic deformation and elasticity

typical of peri-mortem trauma, which could be related to the assassination. The absence of damage on the rib cage allowed the identification of the first thrust that "treacherously struck him in the chest," in agreement with the manservant Strinati's deposition, as the stab wounded Giuliano's right scapula. Moreover, a slant lesion on the ankle cut both fibula and tibia, moving upward from the ventral to the dorsal side. This confirms that he tried to run to the sacristy, when he was struck by a blow that caused him to fall to the ground.

When the victim slumped, the aggressor hit him several times on the same side as indicated by the location of all the wounds; evidence of desperate attempts of defense are attested to by cut marks on the back side of Giuliano's right elbow.

After the reconstruction of the crime dynamic, based on historical testimonies compared with physical pieces of evidence (X-rays, exhumations reports, and pictures and casts of the bones), theories will be presented on the typology of the weapons used, excluding the use of an axe and assuming instead a quite short and heavy weapon. In conclusion, the study demonstrates how a modern forensic approach can increase historical data regarding events and murders of more than five hundred years ago.

Forensic Anthropology, Cold Case, History of Medicine

LW2 The Search for an mtDNA Reference for Abraham Lincoln: Who Was Nancy Hanks?

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After attending this presentation, attendees will learn about the critical role forensic genealogy plays in investigating bio-historical mysteries and the challenges that arise in applying new methodology to investigate a person, in this case Abraham Lincoln, who has been exhaustively researched through conventional means.

This presentation will impact the forensic science community by showing how, as one of the first bio-historical investigations, the study of Abraham Lincoln's genome established a precedent regarding how such investigations will be conducted in the future. The project also reveals fascinating new information about Lincoln and resolves long-standing questions about his ancestry.

It has been suspected that Abraham Lincoln suffered from a variety of genetic disorders including spinocerebellar ataxia, Marfan's syndrome, and the rare cancer MEN2B. However, only recently have ancient DNA analysis techniques become available that could confirm these suspicions that, until now, have been based on Lincoln's physical appearance and historical reports on the condition of his health.

A number of Lincoln assassination relics exist from which

a sample of the President's genome might be obtained for analysis; however, they must first be authenticated through the mitochondrial DNA (mtDNA) obtained from maternally-linked family members. This has proved challenging. It has been difficult to identify and locate Lincoln family members to serve as DNA references. Lincoln has no living descendants, his brother and his sister died without issue, and the family of his mother, Nancy Hanks, has never been conclusively identified. Even if the correct Hanks family is confirmed, it is possible that no match will be found with any relics due to contamination issues or the possibility that Lincoln was adopted or illegitimate. Ongoing efforts are being made to identify suitable relics and obtain permission to use them.

It is known that Nancy Hanks, the President's mother, was from Virginia and resided in Washington County, KY in the late 1700s. There are two leading theories about her parents. According to Adin Baber's *The Hanks Family from Virginia and Westward*, Nancy Hanks was the daughter of Abraham Hanks and Sarah Harper of Virginia. It has also been speculated that the President's mother was the daughter of Lucy Hanks, for many years the only other Hanks living in Washington County. It is interesting that Lucy Hanks appears in Baber's work only as a footnote in the appendix describing uncategorized Hanks families.

MtDNA samples were collected from maternally-linked descendants of Mary Polly Sarah Hanks, a daughter of Abraham Hanks and Sarah Harper, and from maternally-linked descendants of Mary Ann Sparrow and Elizabeth Sparrow, daughters of Lucy Hanks and Henry Sparrow. However, because the mtDNA signature of the two Hanks families matched each other, it was not possible to distinguish between the families. Considering the historical and genealogical aspects of the families, it is highly likely that the mtDNA signature is that of Lincoln's assumed maternal line, even if it has not yet been established which Nancy Hanks, if either, was his assumed mother. The rarity of the Hanks' mtDNA signature also supports this conclusion.

Further autosomal Single-Nucleotide Polymorphism (SNP) tests are planned on Hanks-Harper and Sparrow-Hanks descendants to establish Hanks family structure and to identify where Lincoln's assumed mother, Nancy Hanks, fits within that structure.

Lincoln, Hanks, mtDNA

LW3 The Shame of the Japanese-American Internments

Clarence Moriwaki, BA, 8244 NE Carmella Lane, Bainbridge Island, WA 98110*

After attending this presentation, attendees will be given a solid basis to prevent jumping to unwarranted conclusions at the governmental level concerning the patriotism of Americans of different ethnic or racial grouping.

This presentation will impact the forensic science community by demonstrating the terrible impact on the lives and livelihoods of those forced to endure untold sacrifices without proof of their complicity in anti-American activities except by racial classifications.

It was a sad and even shameful day when Franklin D. Roosevelt's Executive Order resulted in the removal of Japanese-Americans from their homes and livelihoods, mainly on the West Coast of the United States, and their internment in unfamiliar and faraway places in the arid and unfriendly western regions of this country. For the legal system and constitutional law in particular, it was the decision of the United States Supreme Court in 1944 in *Korematsu vs. United States*, 323 U.S. 214, that sustained the legitimacy of the issuance of the Executive Order which has, over

the years, borne the brunt of criticism for upholding the displacement and internment of so many Japanese-Americans. This presentation will provide a short course in these displacements and internments with emphasis on the visual and very graphic panels displaying the heartwrenching plight of the displaced Japanese-Americans which are located in the out-of-doors across the bay on Bainbridge Island.

This presentation will, of necessity, discourse on the issues of national security which prompted the Executive Order at the commencement of WWII as well as whether that occasion may not be afoot with other nationalities even today. Visuals of the somber outdoor panels on Bainbridge Island will enhance the presentation.

Racial Grouping, Anti-American, Ethnic Grouping

LW4 Ceely Rose: Murder in Pleasant Valley

Stewart D. Ryckman, MD, 1468 Brookpark Drive, Mansfield, OH 44906; and Debi Spencer, MFS, CMR 480, BOX 2207, APO, AE 09128*

The goal of this presentation is to introduce attendees to historical facts of rural Richland County, Ohio, interwoven with ghost stories about a triple familicide in 1896, The Ohio State Reformatory, Malabar Farm, author Louis Bromfield, and the movie *The Shawshank Redemption*.

This presentation will impact the forensic science community by examining historical events that include an intriguing murder with ties to paranormal societies and the world of entertainment.

Richland County, Ohio, founded in 1808, has a history encompassing Indian conflicts, Civil War encampments, underground railway stops, and aviation pioneers. The Ohio State Reformatory, open from 1898 to 1990, now only houses ghosts. It has been featured on the TV show *Ghost Adventures*, is listed on many compilations of America's haunted places, and was the site of the filming of *The Shawshank Redemption*. Now a tourist attraction, it is a favorite of paranormal enthusiasts.¹ In Pleasant Valley sits Malabar Farm, home of Louis Bromfield, Pulitzer Prize-winning author of 30 books (four of which were made into movies). Bromfield hosted many Hollywood friends there including James Cagney, Errol Flynn, and Shirley Temple. Humphrey Bogart and Lauren Bacall were married there in 1945. Malabar Farm shares a couple of things with the Reformatory. Although most of the *The Shawshank Redemption* was filmed at the Reformatory, several scenes were filmed near Malabar Farm. The opening scene was filmed at a cabin on the farm. The large, beautiful oak tree where Andy hid the stash and instructions for Red sits just across the road from Malabar. But Malabar Farm also shares the fact that it has its own murder story, which occurred in 1896, the year of Bromfield's birth and just prior to the Reformatory's construction. That story is the focus of this presentation.

Ceely Rose was a troubled girl, a slow learner who today would be classified as developmentally disabled.² She attended school with younger children and was mocked and bullied. She struggled in school and never graduated. By the time she was in her 20s, she became infatuated with neighbor Guy Berry. He was kind to her, but did not reciprocate her feelings. In 1896, she began to tell people that the two of them were going to be married. Guy told her that her family did not approve of him. Ceely decided to kill her family. She mixed arsenic rat poison in the breakfasts of her parents and brother. Her father and brother died. Her mother became ill but recovered. Her mother realized what Ceely had done and tried to protect her by coaching her on what to tell investigators.³ But when her mother began talking about leaving the valley, Ceely became upset, re-poisoned her, and her mother died.

Unable to prove their case, investigators enlisted a friend of Ceely's to trick her into confessing, and Ceely was arrested. After three long trials, she was found not guilty by reason of insanity, became a ward of the state, and was placed in a mental asylum in Toledo, Ohio. Transferred to the Lima State Hospital in 1915, she died there and was buried in 1934 at the age of 53. Her family is interred in Pleasant Valley Cemetery, as is Guy Berry. Richland County is now a frequent destination for paranormal aficionados, as the ghosts of Ceely and her murdered family are said to haunt various sites in Pleasant Valley.⁴ The Reformatory is listed as one of the "scariest places in Ohio." Both locations can be visited by following the Richland County Chamber of Commerce's "Shawshank Trail," which features sites where the movie was filmed.⁵ Ceely's story has been made into both a play and a book.^{6,7}

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Ceely Rose, Ohio State Reformatory, Malabar Farm

LW5 Losing Your Hair in Missouri: Scalping in the American Civil War

Thomas P. Quinn, JD*, 355 S Teller Street, Ste 200, Lakewood, CO 80226

After attending this presentation, attendees will have a better understanding of the prevalence of atrocities committed in the Trans-Mississippi theater of the American Civil War and will learn that scalping was a common practice among Missouri guerillas.

This presentation will impact the forensic science community by educating attendees on a little-known aspect of the American Civil War.

The guerilla warfare on the frontier of Kansas and Missouri during the American Civil War was often vicious and bloody. One of the most brutal pro-confederate guerilla leaders in the Civil War was William T. "Bloody Bill" Anderson who led a band of guerillas targeting Union loyalists and Federal soldiers in Missouri and Kansas. Anderson and some of his men adopted the practice of taking scalps as trophies.

In the autumn of 1864, Anderson and his guerilla company took part in the invasion of northern Missouri by General Sterling Price and his Missouri State Guard. The strategic objective of the offensive was to influence the 1864 presidential election by capturing St. Louis and the state capital of Jefferson City, Missouri. As part of his strategy, Price encouraged guerilla warfare, particularly the disruption of railroads.

On September 27, 1864, Anderson, with a force of about 80 guerillas, many dressed in stolen blue Union uniforms, entered Centralia, Missouri, to cut the Northern Missouri railroad. Riding with Anderson were future outlaws Frank and Jesse James. Anderson's men looted the town and succeeded in blocking the rail line as a train approached carrying civilians and Union soldiers. There were a total of 23 Union soldiers on board who were going home on leave. The guerillas shot dead 22 of the unarmed Union soldiers on the spot then scalped them. They then set fire to the train and sent it running down the tracks.

Following these acts, later on the same day, Anderson's men were pursued by a Union detachment of 155 men from the newly formed 39th Missouri Infantry Regiment. This Union force attempted to fight the mounted and well-armed forces under Anderson in an open battle. The Federal recruits were no match for the heavily armed guerillas. The guerillas shot down 123 of the 155 Union soldiers, many of them while they were attempting to run away or surrender. Afterwards, Anderson's men scalped and mutilated the bodies of many of the dead. There is also evidence some were tortured while still alive.

The legal authority for formation of guerilla units during the Civil War was the Partisan Ranger Act passed by the Confederate Congress in 1862. The purpose of the act was to serve as a stimulus for recruitment of irregular troops into the service of the Confederate Army. While it was later repealed in February of 1864, the Partisan Ranger Act allowed formation of irregular guerilla units in Missouri such as those led by Bloody Bill Anderson and William Quantrill.

None of the guerillas who participated in the Centralia Massacre were charged with war crimes for killing and scalping unarmed Union soldiers; however, many met violent ends during the war and in its aftermath. A few weeks after the Centralia Massacre, Bill Anderson was himself killed with several of his men when they were lured into an ambush by Federal soldiers near Richmond, Missouri. Anderson's body was photographed and dragged through the streets of Richmond.

Scalping, Civil War, Missouri

LW6 Risk Factors for Homicide Victimization in Rwanda, Post-Genocide: A Forensic Epidemiological Investigation

Wilson Rubanzana, MS*, Kacyiru Police Hospital, PO Box 6183, Kigali, Rwanda; Michael Freeman, MD, PhD, 425 NW 10th Avenue, Ste 306, Portland, OR 97209; and Bethany Hedt-Gauthier, PhD, Harvard Medical School, Boston, MA

After attending this presentation, attendees will learn that, despite difficulties in conducting research on sensitive subjects such as homicide in developing countries, field-based forensic epidemiology (collaboration between public health officials and law enforcement agencies in investigating crimes) can be very useful.

This presentation will impact the forensic science community by demonstrating the contribution of forensic epidemiological population-based research in crime investigation in developing countries.

Objective: The study, which is a part of a series of epidemiological investigations, was undertaken to identify homicide victims' characteristics and assess risk factors for violent death in Rwanda, nearly 20 years after the 1994 genocide that took the lives of one million innocent people.^{1,2}

Methods: The investigation relied on the close collaboration between public health researchers and law enforcement services in identifying homicide victims and data

collection.³ The population-based, case-control study was performed from May 2011 to May 2013. One hundred fifty-six cases of homicide victims were investigated from throughout Rwanda. The police investigator provided crime scene information while next of kin supplied socioeconomic and background data. For controls, 468 living study participants, matched for sex, 5-year age group, and village of residence were selected. Next of kin controls were identified and interviewed for control socioeconomic and background data.

Results: Of the 156 homicide victims, 57% were males and 43% were females, and the mean age was 38.2 years (SD=14.9). Thirty-four percent were killed by acquaintances, 28% by intimate partner and 27% by a family member. The patterns of homicide victims show that 81 (51.9%) of homicides occurred inside the victim's home, 53 (34%) in the neighborhood (village of residence), and 22 (14.1%) took place in a relatively remote area. The great majority of victims, 145 (94.2%), underwent a postmortem examination. The most common mechanisms of death were wounds inflicted by sharp instruments (41%, e.g., knives and machetes), followed by blunt force injuries with 36.5%. Of these, 133 (85.3%) of the cases were rural area residents and 23 (14.7%) were urban city dwellers.

Conditional logistic regression analysis indicated that polygamy (OR=7.1), drinking alcohol behavior (slightly: OR=3.2, moderately: OR=5.6, heavily: OR=7.0), previous physical or sexual abuse (OR=4.2), and drug use (OR=11.1) were associated with a high risk of being killed ($p<0.05$). Attaining a secondary level of education (OR=0.2) and being a Christian (OR=0.2) showed protective effects on homicide victimization in Rwanda ($p<0.05$). There was no significant association between being a survivor of genocide, a genocide perpetrator, or a witness in a genocide trial with homicide victimization in Rwanda.

Conclusion: Homicide victims are young and the proportion of female victims is one of the highest in the world. Nearly 20 years after the genocide, homicide victimization risk factors are not unique and specific to Rwanda. Sensitizing communities against polygamy, alcohol and drug abuse, gender-based violence while promoting education, and better understanding the protective link between Christianity and homicide could help protect Rwandan people against homicide deaths.

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Homicide Victimization, Rwanda, Genocide

LW7 Recomposer of the Decomposed: The Career and Last Case of Forensic Sculptor Frank Bender

Thomas A. Crist, PhD, Utica College, 1600 Burrstone Road, Utica, NY 13502; and Zachary R. Lysek, BA*, Northampton County Coroner's Office, 146 Country Club Road, Easton, PA 18045*

After attending this presentation, attendees will understand facial approximation as a tool in forensic identification and the

unique career of forensic sculptor Frank Bender, best known for his age-progressed bust of 18-year fugitive John List and his important contributions to numerous other cold cases.

This presentation will impact the forensic science community by highlighting the use of facial approximation as applied by one of the field's most successful practitioners.

This presentation describes the career and last case of renowned forensic sculptor Frank Bender of Philadelphia (1941-2011), co-founder of the Vidocq Society and self-proclaimed "Recomposer of the Decomposed."

For more than a century, forensic facial approximations or reconstructions have played an important role in identifying decomposed human remains, particularly those from long-unsolved "cold cases." Although facial reconstructions do not meet the *Daubert* standards for positive identification, they are often the most compelling, and successful, components in resolving missing persons cases.

In 1977, commercial photographer and artist Francis Augustus "Frank" Bender visited the Philadelphia Medical Examiner's Office to learn more about human anatomy. Then a student at the Philadelphia Academy of Fine Arts, on his first night at the morgue, Bender saw the body of a woman who had been shot in the head three times. Despite her facial injuries, Bender confidently exclaimed, "I know what she looks like," to Dr. Halbert E. Fillinger, Jr., the forensic pathologist who then asked Bender to work with him on the case by creating a bust from the woman's skull. Based on Bender's work, the woman was identified five months later. Frank Bender's first case was solved and a long forensic partnership with Dr. Fillinger and the Philadelphia Medical Examiner's Office began.

Over the next 34 years, Frank Bender was involved in numerous forensic cases, sculpting more than 40 facial reconstructions. Working from a studio in a converted butcher's shop in Philadelphia, he became one of the best-known forensic artists in the country. While never considering himself a forensic scientist, Bender nonetheless enjoyed noticeable success with many of his cases. Among them was the amazing story of John List, a New Jersey accountant who killed his mother, wife, and children in 1971 and then disappeared. In 1989, Bender worked with forensic psychologist Richard Walter to sculpt an age-progressed bust of List that was featured on an episode of a popular true-crime television series. A neighbor recognized the face portrayed by the bust and two weeks later List was arrested in Richmond, Virginia, where he had lived under a false name. He was convicted of the murders in 1990, sentenced to life imprisonment, and died in prison in 2008.

It was also in 1990 that Frank Bender joined Richard Walter and U.S. Customs Service Special Agent William Fleisher to form the now world-renowned Vidocq Society. Named after the 18th-century French detective Eugène François Vidocq, considered the father of modern criminalistics, the Vidocq Society comprises 150 members of all backgrounds who volunteer their time to investigate and solve "cold case" homicides from across the country.

Fortunately, Frank Bender's last case was resolved shortly before he died on July 28, 2011. Ten years before, on December 30, 2001, police recovered skeletal remains in a wooded area near Easton in Northampton County, Pennsylvania. Anthropological analysis indicated that the remains represented a young woman who presented evidence of mixed European and African ancestry. Despite extensive dental work and several skeletal anomalies, this woman remained unidentified through 2010. In early 2011, the Northampton County Coroner commissioned Frank Bender to sculpt a facial reconstruction of the young woman. Bender had received a diagnosis of terminal cancer two years before, and he knew as he worked on this sculpture that it would be his last one. He revealed the bust to the media in April 2011. In late June, just

one month before he died, the Northampton County Coroner visited Bender to personally give him the rewarding news that the skeleton had been positively identified through a DNA match as a woman from New Jersey who disappeared in 1998.

Frank Bender was an unconventional forensic artist, one with no formal training in forensic science or anatomy but who had an uncanny ability to recreate the faces of the people whose skulls were presented to him by investigators with nowhere else to turn. When asked in an interview shortly before he died how he wanted to be remembered, Frank Bender replied simply, "By what I have done trying to help other people." He continues to do so through the ongoing work of the Vidocq Society, one of his most enduring accomplishments.

Facial Reconstruction, Frank Bender, Vidocq Society

LW8 George Pickett's Part in the Pig War

Michael P. Vouri, BA, National Park Service, 650 Mullis St, Friday Harbor, WA 98250*

After attending this presentation, attendees will learn about a portrayal of the real ability to stave off an impending war by peaceful means.

This presentation will impact the forensic science community by increasing knowledge regarding the relations between the United States and Britain just prior to the Civil War.

Civil War Confederate General George Pickett is best known for leading the charge from which so many of his troops died at Gettysburg; however, in his prior activities as a Captain in the U.S. Army, he played a prominent role in the so-called "pig war" on San Juan Island, now in the state of Washington. The possibility of a war in the northwest between the British, who claimed San Juan Island for Canada, and the United States, who had colonized it as a part of the United States mainland, came to the fore when one settler, Lyman Cutlar, shot a pig in 1859 belonging to the Hudson's Bay Company. That shooting set the stage for the United States and the British's competing claims to jurisdiction over the San Juan Islands. Pickett's Company D of the 9th Infantry was detailed to the island to fend off the British and to keep the peace.

Both the British and the Americans under Pickett were cool-headed and committed to peace without bloodshed. That commitment paid dividends which resulted in an arbitration agreement between the contending countries resolving their dispute in November 1872 with the boundary line between Canada and the United States giving ownership to the San Juan Islands to the U.S. It was the diplomacy of Pickett which was largely in play in fending off hostilities in the "Pig War of 1859."

This presentation will be enhanced by glorious pictures of the San Juan Islands and will serve the educational purpose of proving the merit of negotiation to avert warfare as well as being a living memorial to late General George Pickett and his pertinacity in the face of an impending battle.

War, San Juan, George Pickett



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W5
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Merck Millipore (Discussion of Commercial Products or Services) – K10
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