Breakout Sessions - Schedule & Abstracts

Thursday, May 9

PITTSBURGH MAAFS 2024

Scholarship Winners - Grand Station II

1:00 pm **Identification and Quantification of Illicit Drugs in Blood Using Stir Bar Sorptive Extraction and LC-QQQ-MS**

Abigail Noll[1], B.S., Ashley Ebert[2], Sean Fischer[3], and Stephanie J. Wetzel[1], Ph.D.*

[1]Forensic Science & Law Program, Duquesne University [2]R&D-Analytical Chemistry, Oakwood Laboratories, [3]Testing, Analytics, and Physics, Covestro LLC, Pittsburgh, PA, 15205, USA.

1:20 pm **Machete vs. Knife: A Comparative Study in Sharp Force Trauma**

Lauren Asbury - Towson University

1:55 pm **Developing and Evaluating a Method for Complete Collagenase I Digestion of Bos Taurus Bone Samples**

Reilly Price, BA, Ciara Rhodes, BS, and Sarah Seashols-Williams, PhD - Virginia Commonwealth University*

Biology Session - Grand Station II

2:30 pm **Using the Microbiome to Assess the Health Status of the Individuals Recovered from the East Marshall Street Well.**

Arian Karim, Dr. Baneshwar Singh, Dr. Tal Simmons, Dr. Jenise Swall - Virginia Commonwealth University*

2:45 pm **The Reassociation of Highly Degraded Human Remains Found in the East Marshall Street Well Using Insertion/Null Genotyping and Short Tandem Repeats.**

Sierra L. Laveroni, B.S.; Andrea Malchow B.S., Daniela Frausto, M.S., Michelle M. Woo, M.S., Baneshwar Singh Ph.D., Filipa Simao Ph.D., Tal Simmons Ph.D. - Virginia Commonwealth University*

3:00 pm **DAPI Nuclear Hair Root Staining for STR Typing**

Stephanie M. Betts, Ivette A. Espinoza Quiroga, Hajara S. Chaudhry, Maria Lawas, Susannah C. Kehl, Linda M. Otterstatter, Joseph Donfack - FBI - Research and Support Unit*

3:15 pm **A Microfluidic Device to Separate Biological Components from a Cellular Mixture using Dual-Trap Optical Tweezers**

David J. White, B.S.; Brittney Hackworth, M.S.; Samantha Pagel, M.S.; Mackenzie Lally, M.S.; Sarah Seashols-Williams, Ph.D.; Tracey Dawson Green, Ph.D.; Joseph E. Reiner, Ph.D. - Virginia Commonwealth University*

3:30 pm **Different TrueAllele® users, same DNA answer: a multi-center proficiency study**

William Allan, MS; Jennifer Bracamontes, MS; Matthew Legler, BS; Jonathan Perlin, MS; Mark Perlin, PhD, MD, PhD - Cybergenetics*

4:00 pm **Cellular Autofluorescence for the Determination of Time Since Deposition of Blood Samples**

Hannah E. Lamer, BS; Christopher J. Ehrhardt, PhD; Catherine C. Connon, PhD - Virginia Commonwealth University and Susan A. Greenspoon, PhD - Virginia Department of Forensic Science*

4:30 pm **Development of a new (and free) DNA resource to support the forensic DNA typing Community**
Peter M. Vallone Ph.D. - National Institute of Standards and Technology

Chemistry Session - Grand Station I

2:30 pm **Development of 2D NMR Detection and Quantification Method of Opioids and Their Analogs**
Kathryn James - Waynesburg University

2:45 pm **Development and Application of a Validation Package for Seized Drug Analysis using Rapid GC-MS**
Briana Capistran, Elizabeth L. Robinson, Edward Sisco - National Institute of Standards and Technology*

3:00 pm **Break**

3:30 pm **Scientific Working Group for the Analysis of Seized Drugs (SWGDRUG) Update**
Jennifer Bonetti - Virginia Department of Forensic Science

4:00 pm **GCMS Low Energy Ionization to Determine Structural Information on Fentanyl and Nitazene Analogs**
Kirk Lokits - Agilent Technology

4:30 pm **Forensic Applications of FTIR**
Ron Rubinovitz Ph.D. - ThermoFisher Scientific

Physical Evidence Session - Grand Station III-IV

2:30 pm **Examining the Relevance and Admissibility of Neuroimaging Evidence in Psychopathy and Insanity Defense Cases**
Kaitlyn Svencer, B.A., Jane Moriarty, M.A., J.D., David DeMatteo, J.D., Ph.D., Lyndsie Ferrara, Ph.D. - Duquesne University*

3:00 pm **Using An Indirect Personality Assessment to Determine Psychopathy of Timothy McVeigh**
Mackenzie Miller - - Duquesne University

3:15 pm **Bring out Your Dead: The Skeletons in Institutional Closets**
Jay Bow and Kristy Henson - Fairmont State University*

3:45 pm **The Application of Non-Metric Forensic Anthropological Methods to Virtual 3D Cranial Models**
Marion Davidson MS, Carolyn Rando PhD, Ruth Morgan DPhil - University College London*

4:00 pm **Using historical biocultural variables and vitamin D deficiency to assist with osteoprofiles**
Kristy Henson, MS - Fairmont State University/ University of Leicester

4:15 pm **The Effects of Fingerprint Development Techniques on Forensic Cartridge Case Identification**
Sasha Valentino, Missy Meredith, Sarah Varhola, Stephanie J. Wetzel and Lyndsie Ferrara - Duquesne University*

4:45 pm **ESDA Sequencing for Forensic Document Examination: A Case Study**
Khody Detwiler - Lesnevich & Detwiler

Biology Session - Grand Station II

8:00 am **A new serological method to detect hand epidermal cells in cases involving strangulation**
Thilini Chandrasekara - Virginia Commonwealth University

8:15 am **Visualizing and quantifying DNA on cartridge cases**
Paige Kibler - Virginia Commonwealth University

8:30 am **Getting more from less: low-level DNA mixtures on cartridges**
Kari R. Danser MS, Matthew M. Legler, Megan M. Foley MS, Jennifer M. Bracamontes MS , Mark W. Perlin PhD, MD, PhD - Cybergenetics*

9:00 am **Defeating opposition experts: winning with science**
Jennifer M. Bracamontes, MS, William P. Allan, MS, and Mark W. Perlin, PhD, MD, PhD - Cybergenetics*

9:30 am **An Evaluation of Four Commercial mtDNA Whole Genome Sequencing Kits on Forensic Samples**
Madalynn Swaltek, Courtney Cavagnino, Charla Marshall, and Kimberly Andreaggi - Armed Forces DNA Identification Laboratory/SNA International*

9:45 am **Break**

10:00 am **DNA Identification of Unknown World War II Remains from the Tokyo Prison Fire**
Nickolas Walker, MSFS (AFMES/AFDIL, SNA International), Chelsea Timmerman, MSFS (AFMES/AFDIL, Amentum), Dr. Kristen Grow (Joint Defense POW-MIA Accounting Agency)*

10:30 am **Optimized Recovery of DNA and subsequent STR profiling of different tissues sampled from embalmed human cadavers**
Kofi Afrifah - Bowie State University

11:00 am **New Standards and Best Practices In Biology/DNA**
Charlotte Word - Charlotte J Word

11:30 am **Can the Use of Sharpies in Forensic Analysis be a Source of DNA Transfer and Contamination When Examining Different Scenarios?**
Haley Murphy B.A.; Lyndsie Ferrara, Ph.D.; Julie Sikorsky, M.S.; Pamela Marshall, Ph.D. - Duquesne University*

Interdisciplinary Session - Grand Station I

8:30 am **NIJ funding targeted at public crime laboratories - A historical look at the last eight years**
Tracey Johnson - National Institute of Justice

9:00 am **NamUs: What is it and, Why should your Lab know about it?**
Chuck Heurich - National Institute of Justice

9:15 am **Break**

Chemistry Session - Grand Station I

9:30 am **The use of Leaf Spray Ionization Mass Spectrometry for the Detection of Kratom (Mitragyna speciosa) Leaf**
Macenzie Powell, B.A.; Theodore Corcovilos, Ph.D.; Stephanie Wetzel, Ph.D.; Hannah Zimmerman-Federle, MS; Michael Van Stipdonk, Ph.D - Duquesne University*

9:45 am **Identification and Quantification of Illict Drugs in Blood Using Stir Bar Sorptive Extraction and LC-QQQ-MS**
Abigail Noll[1], B.S., Ashley Ebert[2], Sean Fischer[3], and Stephanie J. Wetzel[1], Ph.D.*
[1]Forensic Science & Law Program, Duquesne University [2]R&D-Analytical Chemistry, Oakwood Laboratories, [3]Testing, Analytics, and Physics, Covestro LLC, Pittsburgh, PA, 15205, USA.

10:00 am **Methods of Application for Internal Standards in Solid-Phase Extraction, in Preparation for Drug Quantitation on LC-QQQ-MS**
Amy E Cook, B.S.; Colette Salerno, M.S.; Pamela Marshall, Ph.D.; Stephanie J Wetzel, Ph.D. - - Duquesne University*

10:15 am **Break**

10:30 am **Break-out Discussion Sessions**

Physical Evidence Session - Grand Station III-IV

9:30 am **Experts Wanted: Demystifying the Student Research Process**
Lyndsie Ferrara - Duquesne University

9:45 am **Using VR in Crime Scene Education, Training, & Proficiency Testing**
Dr. Susan Blankenship, ABC-GKE - University of Maryland Global Campus

10:00 am **The Making of a High-Profile Case: How Media Bias Influences Forensic Investigations in**

Missing Person Cases
Jennifer Fertel - Duquesne University

10:30 am **A Landscape Study: Examining Trends in Serial Killers Raised by Non-biological Parents**
Amanda Piccirilli - Duquesne University

11:00 am **Conducting A Psychiatric Analysis using Collateral Materials: A Case Study of Theodore Kaczynski**
Rebekka Range, B.A*., Lyndsie Ferrara Ph.D., Hannah Stokes Ph.D., John Cencich, J.S.D, and Pamela Marshall, Ph.D. - *Duquesne University*

11:15 am **Identification of Biomarkers Associated with Prolonged Starvation in Cat (Felis catus) Bones**
Annagrace Radocaj B.A., Lisa Ludvico, Ph.D., Becky Morrow DVM, Lyndsie Ferrara, Ph.D., Michael Cascio Ph.D. - Duquesne University*

11:30 am **The Significance of 3D Printed Firearms with Regards to Lethal Capacities and Traceable Elements**
Caitlin Baker, B.A., Zara Ellen Wenzinger, M.S., Stephanie Wetzel, Ph.D., John Viator, Ph.D., Allison Laneve, M.S., Brian Kohlhepp, Pamela Marshall, Ph.D.*

Scholarship Winner Abstracts

Identification and Quantification of Illict Drugs in Blood Using Stir Bar Sorptive Extraction and LC-QQQ-MS

Abigail Noll[1], B.S., Ashley Ebert[2], Sean Fischer[3], and Stephanie J. Wetzel[1], Ph.D.*
[1]Forensic Science & Law Program, Duquesne University [2]R&D-Analytical Chemistry, Oakwood Laboratories, [3]Testing, Analytics, and Physics, Covestro LLC, Pittsburgh, PA, 15205, USA.

The danger proposed by illicit drug use has led to further research into more sensitive substance detection and identification techniques for biological samples. In this study, polydimethylsiloxane-coated magnetic stir bars were used to extract analytes correlating to a fifteen-drug panel from blood samples. The drug panel included illicit substances such as fentanyl and heroin and medical prescriptions like oxycodone and methadone. LC-QQQ-MS with a biphenyl column was used for separation and quantification of these analytes. There have been promising results that the addition of salts has aided the extraction process by making the PDMS more favorable for the drug analytes.

Machete vs. Knife: A Comparative Study in Sharp Force Trauma

Lauren Asbury - Towson University

Differentiating cut marks and sharp force trauma on bone is a crucial component of weapon identification. In an experimental study utilizing three varying motions (hack, stab, slash), I endeavored to see if the weapons' cut marks were still identifiable and unique through varied usage. A hack, stab, and slash each represent a wide array of motions and provide ample opportunity for possible variance between a knife and a machete. In comparing the yield of each weapon on bone following the process of maceration, I found that cut marks were still distinguishable despite their variance. However, at times their identifiable traits overlapped, and thus complicate the process of identification and distinction of weapons.

Developing and Evaluating a Method for Complete Collagenase I Digestion of Bos Taurus Bone Samples

Reilly Price, BA, Ciara Rhodes, BS, and Sarah Seashols-Williams, PhD - Virginia Commonwealth University*

Traditional methods for processing forensic bone samples involve pulverization of bone samples to achieve adequate exposure of genetic material to the extraction buffer. However, this technique can introduce numerous polymerase chain reaction inhibitors, including those found in the extracellular matrix found in mature bone and competition from excessive microbial DNA, in addition to the degradation from pulverization that results in further damage to already degraded genetic material. This project proposes using demineralization with complete collagenase I digestion of bone slices as a method for improving the quality and quantity of DNA obtained from forensic bone samples while preventing the incorporation of PCR inhibitors. Through the use of fluorescence microscopy, quantitative PCR, and short tandem repeat analysis, the digestion protocol was optimized, and the impact of demineralization and collagenase I digestion on DNA was assessed. While collagenase I was found to have deleterious effects on DNA yield and STR profile quality, demineralization of bone slices produced comparable DNA yields and improved profile quality compared to pulverized control samples.

Interdisciplinary Session Abstracts

NIJ funding targeted at public crime laboratories - A historical look at the last eight years
Tracey Johnson - National Institute of Justice

Starting in 2015, NIJ initiated an annual solicitation specifically aimed at providing funding to public forensic laboratories titled Research and Evaluation for the Testing and Interpretation of Physical Evidence in Publicly Funded Forensic Laboratories. The goal of this solicitation is to support research to evaluate current laboratory methods or emerging methods. Awards under this solicitation produce deliverables that include best practices to improve efficiency, accuracy, reliability, and cost-effectiveness as well as protocols that can be adopted by the community. Over the last eight years, this solicitation has provided funding for thirty-eight awards across multiple disciplines including DNA, impression and pattern evidence, seized drugs, and toxicology. Historic output from these awards will be highlighted as part of presentation. In addition, NIJ will recommend and provide resources to encourage public laboratories engagement with research and contributions toward building a positive research culture within the forensic sciences. NIJ will explain how these activities can assure improvements to both scientific integrity and quality within the forensic disciplines.

NamUs: What is it and, Why should your Lab know about it?
Chuck Heurich - National Institute of Justice

NamUs is a nationwide, centralized, repository for missing, unidentified, and unclaimed persons cases. The core is the database but, the program offers free forensic, investigative, and analytical services. This presentation will give an overview of NamUs (with updates) and discuss why crime labs should be aware of the program.

Biology Session Abstracts

Using the Microbiome to Assess the Health Status of the Individuals Recovered from the East Marshall Street Well.

Arian Karim, Dr. Baneshwar Singh, Dr. Tal Simmons, Dr. Jenise Swall - Virginia Commonwealth University*

In 1994, during the construction of the Kontos Building on the Medical College of Virginia Campus, a well on East Marshall Street was discovered to contain artifacts and human remains of African descent. Upon archaeological analysis by the Smithsonian, the ancestral remains can be dated back to the mid-19th century, when grave robbing and the use of cadavers of African descent in medical schools were common. The aims of this project is to use dental calculus recovered from the mandible and maxilla to gain insight into the ancestral remains health environment, and to compare modern and ancient dental calculus to determine differences in the oral microflora over the past few centuries through 16S rDNA sequencing of the V4 region. The taxonomy profiles revealed the presence of pathogenic bacteria that can be attributed to disease and major differences between the dental calculus of the East Marshall Street Well Individuals and modern dental calculus. Most notably, the bacteria known to cause Tetanus (Clostridium tetani), and Tuberculosis (Mycobacterium tuberculosis) were found in a very high abundance in three individuals recovered from the East Marshall Street Well.

The Reassociation of Highly Degraded Human Remains Found in the East Marshall Street Well Using Insertion/Null Genotyping and Short Tandem Repeats.

Sierra L. Laveroni, B.S.; Andrea Malchow B.S., Daniela Frausto, M.S., Michelle M. Woo, M.S., Baneshwar Singh Ph.D., Filipa Simao Ph.D., Tal Simmons Ph.D. - Virginia Commonwealth University*

The East Marshall Street Well (EMSW) contained a minimum of 53 commingled individuals, artifacts, and other historical items dating to the mid-19th century. All 572 of the human limb bones from the well were analyzed using the InnoQuant® HY and InnoTyper® 21 Human DNA Analysis kits (InnoGenomics, New Orleans, LA) for Insertion/Null genotypes. Two-hundred and fifty samples had a quantitation value of >0 .025 ng and, of those sampled, 143 displayed either a full or partial profile following amplification and detection. These samples were then re-associated into 16 discrete partial individuals using the Familias Software. Short tandem repeat analysis is on-going in attempts to re-associate crania and mandible elements to long bone groups.

DAPI Nuclear Hair Root Staining for STR Typing

Stephanie M. Betts, Ivette A. Espinoza Quiroga, Hajara S. Chaudhry, Maria Lawas, Susannah C. Kehl, Linda M. Otterstatter, Joseph Donfack - FBI - Research and Support Unit*

The DNA binding dye 4', 6-diamidino-2-phenylindole (DAPI) has been shown to be an efficient and cost-effective method for screening hair roots for the presence of visible nuclei. In this study, 183 hair samples were collected and microscopically examined by a qualified Forensic Trace Examiner for their suitability to be routed for nuclear DNA or mitochondrial DNA analysis. The hairs samples were DAPI-stained and further classified into five bins based on the number of visible nuclei detected in hair roots: bin 1 (0 nuclei), bin 2 (1 – 24 nuclei), bin 3 (25 – 49 nuclei), bin 4 (50 – 99 nuclei) and bin 5 (> 100 nuclei). The bins were used to determine the minimum number of hair root nuclei required to yield an STR profile suitable for National DNA Index System (NDIS) upload. Results from this study showed that DNA yield positively correlated with the number of nuclei. In addition, a threshold was identified such that hair samples containing at least 25 visible nuclei could produce STR profiles that are suitable for NDIS upload.

A Microfluidic Device to Separate Biological Components from a Cellular Mixture using Dual-Trap Optical Tweezers

David J. White, B.S.; Brittney Hackworth, M.S.; Samantha Pagel, M.S.; Mackenzie Lally, M.S.; Sarah Seashols-Williams, Ph.D.; Tracey Dawson Green, Ph.D.; Joseph E. Reiner, Ph.D. - Virginia Commonwealth University*

DNA interpretation of biological mixtures, a recurrent consequence of sexual assault-type evidence, is arguably one of the most challenging and time-consuming steps in the forensic DNA analysis workflow, often requiring the use of complex back-end mixture deconvolution procedures. An alternative to the traditional differential extraction method is optical trapping, a non-invasive method that utilizes the properties of light to create an optical trap for cell separation and manipulation. The optical trap is created by tightly focusing a laser beam through a high-numerical immersion objective on an inverted microscope, where the gradient force applied by the laser allows for individual cells or cell clusters (e.g., spermatozoa, vaginal epithelial cells, and leukocytes) to become captured and fixed to a high-intensity region; through the use of a joystick, the cells are manipulated in the X, Y, and Z axes. Previous work has demonstrated successful isolation and complete, single-source STR profiles by trapping 15 leukocytes or 40 sperm cells, with minimal carryover from the female epithelial donor when evaluating mock sexual assault evidence. Recent work has focused on optimizing optical trapping in a closed microfluidic device system, which generates equivalent results and has reduced contamination potential. Optical trapping has the potential to provide superior separation compared to that of differential extraction, and ultimately, could reduce the labor and time required by the analyst in an attempt to eliminate mixtures at the beginning of the DNA analysis workflow.

Different TrueAllele® users, same DNA answer: a multi-center proficiency study

William Allan, MS; Jennifer Bracamontes, MS; Matthew Legler, BS; Jonathan Perlin, MS; Mark Perlin, PhD, MD, PhD - Cybergenetics*

The objective TrueAllele® genotyping computer gets the same DNA match statistics, regardless of laboratory or analyst. The identification information doesn't depend on sequencer or STR kit. TrueAllele learns lab parameters from evidence data without calibration. Our multi-center study shows that analysts everywhere get everything at once from all their DNA data.

Cellular Autofluorescence for the Determination of Time Since Deposition of Blood Samples

Hannah E. Lamer, BS; Christopher J. Ehrhardt, PhD; Catherine C. Connon, PhD - Virginia Commonwealth University and Susan A. Greenspoon, PhD - Virginia Department of Forensic Science*

An estimate of the time biological material was deposited at a crime scene, referred to as the time since deposition (TSD), would provide powerful information for forensic cases. This study investigated TSD of bloodstains over 180 days via cellular autofluorescence measured using flow cytometry. Results showed that there is no significant difference in average autofluorescence over a period of 180 days, however detectable differences in green autofluorescence between blood and other biological source types were observed through a TSD of 30 days. In addition, there is a positive correlation between average frequency of cells with autofluorescence intensity greater than 104 and TSD. These results suggest that average fluorescent intensity might not be used to estimated TSD for bloodstains, however questions remain such as whether the autofluorescence of RBCs at multiple wavelengths is obscuring the TSD autofluorescent pattern of white blood cells.

Development of a new (and free) DNA resource to support the forensic DNA typing Community
Peter M. Vallone Ph.D. - National Institute of Standards and Technology

Given challenges with developing DNA reference materials and recognizing the need for biological samples in validation studies, NIST has developed a new classification of exploratory material called a “Research Grade Test Material (RGTM).” RGTMs aim to evaluate fit-for-purpose needs within a community, and to this end, we are developing RGTM 10235 - Forensic DNA Typing Resource Material. The material is composed of a set of eight well-quantified DNA extracts. Components include three single source samples, two degraded samples, and three mixture samples. The preparation of the material and resulting quantification and CE typing data from participating labs will be presented.

A new serological method to detect hand epidermal cells in cases involving strangulation
Thilini Chandrasekara - Virginia Commonwealth University

In cases of assaults involving strangulation, methods are needed that can detect the presence of hand epidermal cells and differentiate them from other types of skin epidermal cells that are derived from the victim’s neck. The aim of this study was to develop a new front-end method for screening biological samples for the presence of hand epidermal cells based on their morphological and autofluorescence characteristics. Results showed that hand epidermal cells were detected in 10 different mock mixture samples comprised of multiple types of skin cells and can be integrated within the operational workflow of a DNA caseworking unit. This has the potential to be utilized as a front-end analysis technique prior to DNA analysis and also provide probative information for a variety of assault cases.

Visualizing and quantifying DNA on cartridge cases
Paige Kibler - Virginia Commonwealth University

How much DNA is left on cartridge cases after the firing process and how can these evidence samples be processed has been a current question for forensic laboratories. Besides having trace amounts of DNA originally, PCR inhibitors and degradation from the firing process are two other issues when dealing with this type of evidence. A common solution for removing inhibitors is the rinse solution which chelates metal ions such as copper. Samples have been rinsed, visualized, and quantified to determine how much DNA is present and lost during the firing process.

Getting more from less: low-level DNA mixtures on cartridges
Kari R. Danser MS, Matthew M. Legler, Megan M. Foley MS, Jennifer M. Bracamontes MS , Mark W. Perlin PhD, MD, PhD - Cybergenetics*

How much identification information can be recovered from firearm cartridges? Our study examined DNA data from different casing materials and collection methods. On the same STR data, we compared TrueAllele® computer interpretation with simple allele counting. TrueAllele measured more information and found previously unidentified contributors.

Defeating opposition experts: winning with science
Jennifer M. Bracamontes, MS, William P. Allan, MS, and Mark W. Perlin, PhD, MD, PhD - Cybergenetics*

An opposition expert's argument may confuse a judge or jury. In a recent criminal case, the DNA opponent undermined probabilistic genotyping error rates. They misread a published validation study to incorrectly find a high error rate. This talk shows our successful response to the flawed attack.

An Evaluation of Four Commercial mtDNA Whole Genome Sequencing Kits on Forensic Samples
Madalynn Swaltek, Courtney Cavagnino, Charla Marshall, and Kimberly Andreaggi - Armed Forces DNA Identification Laboratory/SNA International*

The practice of massively parallel sequencing (MPS) for mitochondrial DNA can be quite useful in missing persons cases, which involve degraded skeletal remains and distant relatives, as well as in cases involving shed hairs (rootless hair shafts). The use of MPS paired with commercial mitogenome kits make it more feasible for laboratories to adopt mtDNA analysis, and a method comparison involving different mitogenome methodologies can help inform laboratories about the considerations that should be made when generating and analyzing mitogenome data. In this study, 16 samples ranging in quality were processed following the manufacturer's protocol for the following commercial kits: ForenSeq mtDNA Whole Genome Kit, PowerSeq WGM System, NimaGen IDseek Mitochondrial DNA Full Genome Sequencing, and QIASeq Targeted DNA Human Mitochondrial Panel. Data were generated on the MiSeq FGx and analyzed using the CLC Genomics Workbench, Universal Analysis Software, and/or GeneMarker HTS. The mtDNA sequencing results were compared to evaluate the performance of commercially available mtDNA sequencing methods for samples of varying quality.

Disclaimer: The opinions or assertions presented hereafter are the private views of the speaker(s) and should not be construed as official or as reflecting the views of the Department of Defense, its branches, the Defense Health Agency, or the Armed Forces Medical Examiner System. Any mention of commercial products was done for scientific transparency and should not be viewed as endorsement of the product or manufacturer.

DNA Identification of Unknown World War II Remains from the Tokyo Prison Fire
Nickolas Walker, MSFS (AFMES/AFDIL, SNA International), Chelsea Timmerman, MSFS (AFMES/AFDIL, Amentum), Dr. Kristen Grow (Joint Defense POW-MIA Accounting Agency)*

The Tokyo Prison Fire project involves anthropological and DNA analysis of human remains, which were severely burnt during U.S. firebombing air raids performed across Imperial Japan in 1945. Remains recovered from the Tokyo prison were buried as unknowns in the Philippines Manila American Cemetery after the war. In 2022 the unknowns were disinterred and honorably transferred to the Defense POW/MIA Accounting Agency (DPAA) laboratory located at Joint Base Pearl Harbor-Hickam (JBPHH) Hawaii in an effort to identify the unknown service members. Innovative mitochondrial (mtDNA) next-generation sequencing (NGS, aka MPS) and Sanger mtDNA sequencing methods, autosomal short tandem repeat (auSTR) analysis using the AmpFLSTRTM Minifiler and PowerPlex® Fusion kits, and an enhanced detection analysis method for Y-chromosomal Short Tandem Repeat (Y-STR) using the AmpFLSTRTM Yfiler kit were used. AFMES-AFDIL (Armed Forces DNA Identification Laboratory-Armed Forces DNA Identification Laboratory) is attempting to identify these long-lost service members through comparison to servicemember family reference samples.

Disclaimer: The opinions or assertions presented hereafter are the private views of the speaker(s) and should not be construed as official or as reflecting the views of the Department of Defense, its branches, the Defense Health Agency, or the Armed Forces Medical Examiner System. Any mention of commercial products was done for scientific transparency and should not be viewed as endorsement of the product or manufacturer.

Optimized Recovery of DNA and subsequent STR profiling of different tissues sampled from embalmed human cadavers

Kofi Afrifah - Bowie State University

Storage of specimen sampled from human remains for pathological testing, embalming for burial purposes, and for human identification often requires formalin fixation and/or paraffin embedding. This research sought to extract amplifiable DNA from thirteen brain, bone marrow and cartilage samples from four formalin embalmed human cadavers. Brain, cartilage and bone marrow samples were taken from four different cadavers at autopsy at the Ghana Police Hospital mortuary in Accra, Ghana sixty-two days after embalming. An optimized preparation and DNA extraction protocol was carried out on all the samples. Brain samples were also taken from a non-formalin treated fifth cadaver of known STR profile, and standard DNA extraction performed to serve as positive control. Our optimized protocol yielded detectable quantities of DNA from the samples when quantified with the 7500 Real-Time PCR equipment. The extracted DNA also yielded full STR profiles with varying peak heights for forensic identification purposes. The measured degradation indexes of the DNA samples were greater than 1.0, with peak heights of generated STR profiles above the limits of detection of the 3500 genetic analyzer.

Can the Use of Sharpies in Forensic Analysis be a Source of DNA Transfer and Contamination When Examining Different Scenarios?

Haley Murphy B.A.; Lyndsie Ferrara, Ph.D.; Julie Sikorsky, M.S.; Pamela Marshall, Ph.D. - Duquesne University*

Body fluids on fabric items are a commonly encountered type of evidence at crime scenes. During evidence processing, the fluid is outlined using a Sharpie® marker to assist DNA analysts to sample the correct area. The tip of the Sharpie®, which has direct contact with the fabric, is not decontaminated between uses. Additionally, the felt fibers of a Sharpie® tip are similar to the natural fibers of a cotton swab, which are utilized in crime labs to collect sources of DNA. The similarities between swab and Sharpie® fibers as well as repeated direct contact with body fluid-stained evidence increases the potential for DNA transfer. The ability of a Sharpie® to collect DNA was compared using an accidental sampling scenario and mock casework scenario.

New Standards and Best Practices In Biology/DNA

Charlotte Word - Charlotte J Word

Several new standards and best practice recommendations drafted by the Organization of Scientific Area Committees for Forensic Science (OSAC) and developed by the AAFS Academy Standards Board (ASB) have become available recently for implementation in forensic laboratories conducting DNA testing. Information regarding some of the new standards will be presented. Featured documents will include:

1. ANSI/ASB Standard 123, Standard for Routine Internal Evaluation of a Laboratory's DNA Interpretation and Comparison Protocol, First Edition, 2024
2. ANSI/ASB Standard 139, Reporting DNA Conclusions, First Edition, 2024
3. ANSI/ASB Standard 154, Standard for Training on Testimony for Forensic Biology, First Edition, 2024
4. ANSI/ASB Best Practice Recommendation 171, Best Practice Recommendations for the Management and Use of Quality Assurance DNA Elimination Databases in Forensic DNA Analysis, First Edition, 2024

General information regarding OSAC and ASB will also be presented.

Chemistry Session Abstracts

Development of 2D NMR Detection and Quantification Method of Opioids and Their Analogs
Kathryn James - Waynesburg University

Fentanyl is an illicit drug that has quickly become the dominant opioid in Pennsylvania, contributing to the death of fifteen people per day in 2021. The number of overdose deaths has continued to climb as DEA analysts discover that fentanyl tablets frequently contain a dose 2.5 times above the lethal dose. Currently, crime laboratories use GC to detect the presence of fentanyl. However, due to the emergence of fentanyl analogs, the corresponding change in retention time renders chromatographic methods of detection and identification ineffective. We have set out to develop an NMR-based method to detect and quantify fentanyl and its analogs using 2D NMR techniques, which makes the method more robust in the presence of other analytes that crowd the spectrum.

Development and Application of a Validation Package for Seized Drug Analysis using Rapid GC-MS
Briana Capistran, Elizabeth L. Robinson, Edward Sisco - National Institute of Standards and Technology*

With the lack of standardized validation protocols that exist among the forensic community, validating new methods and technologies can be a challenging, yet necessary, task. This presentation will highlight a validation package developed for rapid GC-MS, a screening technique increasingly being implemented in forensic laboratories. The package includes a full validation plan and accompanying workbook for seized drug applications, and it is designed as a freely available template for use in practicing laboratories to ultimately lower the barrier of implementation. Validation of an in-house rapid GC-MS system was conducted according to the plan and demonstrated adequate instrument performance for seized drug analysis and identified limitations of the technique (e.g., isomer differentiation). Additional work is currently underway to extend the validation package to other areas of forensic analysis, such as ignitable liquid analysis.

Scientific Working Group for the Analysis of Seized Drugs (SWGDRUG) Update
Jennifer Bonetti - Virginia Department of Forensic Science

The Scientific Working Group for the Analysis of Seized Drugs (SWGDRUG) was founded in 1997 with the mission of improving the quality of the forensic examination of seized drugs and responding to the needs of the forensic community. This is done through developing internationally accepted minimum standards, identifying best practices within the international community, and providing resources to help laboratories meet these standards.

The SWGDRUG core committee is comprised of representation from federal agencies, regional associations, international organizations, as well as academic institutions. This presentation will showcase the resources provided by SWGDRUG and highlight recent updates, on-going developments, and future directions.

GCMS Low Energy Ionization to Determine Structural Information on Fentanyl and Nitazene Analogs
Kirk Lokits - Agilent Technology

This work compiles low energy ionization spectra of fentanyl and nitazene analogs to assist in the identification of isobaric spectra generated under 70 eV ionization energy. Low energy ionization spectra of nitazene fentanyl analog standards were generated at 10, 12, 15, and 17 eV ionization energies and optimized for the formation of a molecular ion, molecular ion abundance, and high spectral fidelity (isotopic ratios) of the molecular ion patterns. The spectral patterns were then compared to street drug case samples and identified nitazene analogs and other controlled substances based on the creation of their respective molecular ion and isotopic patterns.

Forensic Applications of FTIR

Ron Rubinovitz Ph.D. - ThermoFisher Scientific

As a rapid, non-destructive method, FTIR is a well-established tool of the forensic scientist, as it utilizes a variety of specific techniques for a large range of sample types. In this presentation, the advantage of FTIR to quickly, and with minimal sample preparation, identify samples will be illustrated in a review of forensic applications. Forensic studies and advances in IR microscopy instrumentation will be presented, as well as the benefits of expanding the spectral range of FTIR microscopy and attenuated total reflection (ATR) measurements. Also, the advantages of FT-Raman spectroscopy as a complimentary non-destructive identification method and its safety benefits will be discussed. Finally, specific examples of the power of FTIR to differentiate between isomers will be reviewed with respect to cathinones as well as the benefits of combining FTIR with gas chromatography.

The use of Leaf Spray Ionization Mass Spectrometry for the Detection of Kratom (Mitragyna speciosa) Leaf

Macenzie Powell, B.A.; Theodore Corcovilos, Ph.D.; Stephanie Wetzel, Ph.D.; Hannah Zimmerman-Federle, MS; Michael Van Stipdonk, Ph.D - Duquesne University*

Mitragyna speciosa is a plant that produces a metabolite called mitragynine and recently it has become available in the United States and is commonly known as kratom. Kratom is currently not considered an illegal substance in the United States; however, the Drug Enforcement Administration has deemed it a drug of concern. The purpose of this study is to detect Mitragyna speciosa using Leaf spray Ionization Mass Spectroscopy (LSI-MS), this was proven to be successful. Three strains tested, meng da, red meng da, and white meng da and all contained a parent ion peak at 399 m/z which is indicative of the [M+H] peak of kratom as it has a molecular weight of 398.5 g/mol. The fragment ion peaks that were present in the CID scans of 399 were 364m/z, 238m/z, 226m/z, and 174m/z. From these results, the hypothesis stands that kratom can be detected using LSI-MS.

Identification and Quantification of Illict Drugs in Blood Using Stir Bar Sorptive Extraction and LC-QQQ-MS

Abigail Noll[1], B.S., Ashley Ebert[2], Sean Fischer[3], and Stephanie J. Wetzel[1], Ph.D.*
[1]Forensic Science & Law Program, Duquesne University [2]R&D-Analytical Chemistry, Oakwood Laboratories, [3]Testing, Analytics, and Physics, Covestro LLC, Pittsburgh, PA, 15205, USA.

The danger proposed by illicit drug use has led to further research into more sensitive substance detection and identification techniques for biological samples. In this study, polydimethylsiloxane-coated magnetic stir bars were used to extract analytes correlating to a fifteen-drug panel from blood samples. The drug panel included illicit substances such as fentanyl and heroin and medical prescriptions like oxycodone and methadone. LC-QQQ-MS with a biphenyl column was used for separation and quantification of these analytes. There have been promising results that the addition of salts has aided the extraction process by making the PDMS more favorable for the drug analytes.

Methods of Application for Internal Standards in Solid-Phase Extraction, in Preparation for Drug Quantitation on LC-QQQ-MS

Amy E Cook, B.S.; Colette Salerno, M.S.; Pamela Marshall, Ph.D.; Stephanie J Wetzel, Ph.D. - - Duquesne University*

Toxicological analysis requires high precision and accuracy during sample preparation, which involves the extraction of analytes of interest from an often complex biological matrix. One possible technique to increase efficiency is pre-loading deuterated internal standards to later integrate into Solid-Phase Extraction (SPE), cutting out time-consuming steps and possible human error in sample preparation. In this study, SPE was performed on drug-spiked synthetic urine samples using pre-loaded frit materials and SPE cartridges, then Liquid Chromatography–Triple Quadrupole–Mass Spectrometry (LC–QQQ–MS) was used to analyze the extracts and determine percent recovery for each drug in all samples and controls. In addition, a time study was conducted in which the pre-loaded materials were stored to determine if recovery decreased over time. These experiments provide useful evidence about the reliability of pre-loaded internal standards, as well as the potential of creating pre-loaded frits and cartridges to eventually market to laboratories.

Physical Evidence Session Abstracts

Examining the Relevance and Admissibility of Neuroimaging Evidence in Psychopathy and Insanity Defense Cases

Kaitlyn Svencer, B.A., Jane Moriarty, M.A., J.D., David DeMatteo, J.D., Ph.D., Lyndsie Ferrara, Ph.D. - Duquesne University*

Neuroimaging, the visual aspect of neuroscience, refers to various forms of technology that are used to image the brain structurally and functionally. With increased utilization in court, this evidence type has been examined in various applications of the law in terms of relevance and admissibility of brain scans for physical brain trauma. The goal of this research was to expand on that work to assess how brain scans are introduced and applied in various insanity defense and psychopathy cases. The insanity defense portion compared mental health assessment evidence and neuroimaging evidence, while the psychopathy cases analyzed the role of neuroimaging to understand the application of this evidence type. Overall, the research investigated how neuroimaging evidence is applied in court.

Using An Indirect Personality Assessment to Determine Psychopathy of Timothy McVeigh

Mackenzie Miller - Duquesne University

Timothy McVeigh, the Oklahoma City Bomber, will be analyzed using the personality assessment known as an Indirect Personality Assessment. This type of assessment is done to analyze the unconscious motives and feelings of the individual through interrogation of themselves and those around them. McVeigh was executed in June 2001, for the interrogation portion of the assessment, the interview tapes and biography, American Terrorist: Timothy McVeigh and the Oklahoma City Bombing by Lou Michel and Dan Herbeck, will be used. Through this research, McVeigh will be analyzed to determine if he exhibits psychopathic behaviors.

Bring out Your Dead: The Skeletons in Institutional Closets

Jay Bow and Kristy Henson - Fairmont State University*

Human skeletal remains have a long history of use in natural science courses leading many institutions to accumulate collections over time. The majority of these antique teaching skeletons have no records associated with them, making it likely that they were obtained during an ethically gray era and were unassociated. To rehumanize and reassociate the individuals in Fairmont State University's collection, osteometric and macroscopic analysis techniques were used to reunite disarticulated individuals and determine their osteobiography. This has resulted in the discovery that the remains belong to a minimum of 43 individuals, 28 of whom were able to have profile information restored. The current ethical and legal standards in place in the US regarding teaching skeletons are lax, but small-scale efforts by individuals and institutions can work to rectify the decades of immoral treatment of the literal skeletons in institutional closets.

The Application of Non-Metric Forensic Anthropological Methods to Virtual 3D Cranial Models
Marion Davidson MS, Carolyn Rando PhD, Ruth Morgan DPhil - University College London*

Research is exploring the analysis of radiological data as a proxy for real human remains within forensic anthropological methods. This study examined the application of the most practiced cranial non-metric sex and population affinity estimation methods to human skeletal remains and their virtual three-dimensional bone model counterparts created from computed tomography. Participants with osteological experience observed both the real skulls and their 3D model counterparts, recording non-metric traits, sex and population affinity estimations, and their confidence in scoring these parameters. This presentation presents results and key factors surrounding the comparable application of methods and discusses the variables that impacted this experiment, including osteological training, virtual model experience, and the effects of scanning and modelling parameters.

Using historical biocultural variables and vitamin D deficiency to assist with osteoprofiles
Kristy Henson, MS - Fairmont State University/ University of Leicester

The biocultural approach uses biological, environmental, and cultural variables to help identify where an individual spent their childhood or their adult life. This research examines historic skeletal, genealogical, and biocultural information to determine if there is a correlation with the presence of vitamin D deficiency. One hundred seventy-seven individuals living between the 1830s and 1940s were analyzed and traced through the historical record. Preliminary results indicate that there were trends in determining where an individual spent their childhood and the likelihood of vitamin D deficiency (chi-square $p = 0.006$; Spearman RS 0.92, $p = -0.008$), year of death (chi-square 0.005; Spearman RS 0.87, $p = -0.01$), presence of spinal pathology (chi-square $p = 0.05$; Spearman RS 0.72, $p = -0.03$), and trending toward age ($p = 0.08$). More research is warranted but completing osteoprofiles using biocultural variables may help identify where someone is from and how they lived their lives which in turn will assist with identification.

The Effects of Fingerprint Development Techniques on Forensic Cartridge Case Identification
Sasha Valentino, Missy Meredith, Sarah Varhola, Stephanie J. Wetzel and Lyndsie Ferrara - Duquesne University*

The purpose of this research was to determine what effects certain fingerprint development techniques have on cartridge cases and if they impact cartridge case comparisons. Brass, steel and aluminum cartridge cases in both 9mm and .45 calibers were processed using cyanoacrylate fuming, gun blue, basic yellow 40, black powder and a sequence of these techniques. The cartridge cases also were cleaned using acetone, soapy water and alcohol wipes. It was determined that gun blue and the sequence of techniques have a negative effect on the comparisons and all of the cleaning methods aided comparisons.

ESDA Sequencing for Forensic Document Examination: A Case Study
Khody Detwiler - Lesnevich & Detwiler

This presentation investigates the significance of employing the ESDA (Electrostatic Detection Apparatus) sequencing method to determine the chronological order of ink strokes and intersecting handwriting impressions in certain types of forensic document examination cases. The study highlights the pivotal role of ESDA sequencing in unraveling the production sequence of a series of business records in a particular case. Contrary to the assertions made, the findings from ESDA sequencing examination provided critical insights, shedding light on the true chronological sequence of production. This abstract highlights the significance of the ESDA sequencing method as a valuable asset in forensic document analysis, especially in cases where determining production sequencing or document dating is of utmost importance.

Experts Wanted: Demystifying the Student Research Process
Lyndsie Ferrara - Duquesne University

Experts are wanted to assist and enhance student research projects, but many may be unfamiliar with the process. From generating ideas to aiding with design, execution, and ideally publication, practitioners can provide meaningful contributions without the process being burdensome. This presentation explains the student research process and how practitioners can engage with students and universities in a way that is mutually beneficial and more impactful for the forensic science community.

Using VR in Crime Scene Education, Training, & Proficiency Testing
Dr. Susan Blankenship, ABC-GKE - University of Maryland Global Campus

Many people learn by doing, and crime scene investigation is a job where most of the learning is accomplished by doing the tasks rather than reading about them. However, the worst place to learn by doing is on an active crime scene, as crucial mistakes can affect the overall outcome of the justice system. Creating training crime scenes can be time consuming difficult, and inconsistent. This is where virtual reality can help. VR crime scenes can allow for teaching training, and evaluation, without much preparation on the part of the trainer. The actions taken within the VR crime scene can also be recorded, allowing for an after-action debriefing. The VR crime scene and recording can also be used for testing, including proficiency testing, providing a fuller experience for both the tester and test taker than the current experience of testing only individual skill sets or paying for an expensive one time use crime scene. Overall, virtual reality can increase the value to crime scene investigation via education, training, and proficiency testing.

The Making of a High-Profile Case: How Media Bias Influences Forensic Investigations in Missing Person Cases
Jennifer Fertel - Duquesne University

Media bias often prioritizes white women over underrepresented individuals based on race, sexuality, ethnicity, or gender, thus neglecting those who don't fit societal ideals. To examine how media impacts forensic investigations, similar missing person investigations were compared. Additionally, a series of interviews were conducted with forensic investigators and media representatives. Both positive and negative impacts were observed but were found to be case-dependent. However, influences such as community involvement, finances and societal standing were observed in all cases.

A Landscape Study: Examining Trends in Serial Killers Raised by Non-biological Parents
Amanda Piccirilli - Duquesne University

While serial killers can be found everywhere in the world, they became increasingly prevalent in the United States beginning in the 1960s. Using the Radford/Florida Gulf Coast University Serial Killer Database, trends in serial killers who were adopted, placed in foster care, or raised by family members other than their biological parents were examined. Out of 500 serial killers active between 1965 to 2003, 10% were raised by their non-biological parents. This study illustrated trends and numerical data within the population of serial killers raised by non-biological parents. This will be useful to further research on a subset of American serial killers and to provide resources to law enforcement and researchers using the database.

Conducting A Psychiatric Analysis using Collateral Materials: A Case Study of Theodore Kaczynski
Rebekka Range, B.A*., Lyndsie Ferrara Ph.D., Hannah Stokes Ph.D., John Cencich, J.S.D, and Pamela Marshall, Ph.D. - *Duquesne University*

Competency to stand trial and sentencing are both factors impacted by the results of a mental health evaluation. Currently, these evaluations utilize interviews with the defendant to answer the question of their mental state. In cases of Fifth Amendment invocations, these individuals do not have to speak with the mental health expert and a method of analysis that does not require the interview becomes necessary. A case study of Theodore Kaczynski has been used to develop such a method, specifically with documents. These documents are being compared to the diagnostic criteria found in the DSM-5 (2013) as well as the PDM-2 (2008) to determine if a potential diagnosis can be reached. This research will impact the forensic science and legal community by presenting a method to assess mental health disorders when an individual invokes their Fifth Amendment rights and refuses to participate in psychological evaluations.

Identification of Biomarkers Associated with Prolonged Starvation in Cat (Felis catus) Bones
Annagrace Radocaj B.A., Lisa Ludvico, Ph.D., Becky Morrow DVM, Lyndsie Ferrara, Ph.D., Michael Cascio Ph.D. - Duquesne University*

In incidents of hoarding animals, starvation or emaciation of the animal is not uncommon; in the late stages of decomposition, little to no tissue is present, and only depleted bone samples can be collected from the victim. This research aims to investigate the correlation between the N-telopeptide concentrations and starvation through the matrix of bone. Immunoassay testing of starved and non-starved feline serum was performed and analyzed as a proof-of-concept study. Once obtained, the presumably starved and non-starved bone samples were ground into a fine powder, followed by a protein extraction; the extracted samples were then subjected to immunoassay testing. The results indicated that in serum, the N-telopeptide concentration of the starved samples tended to be higher; however, further testing will be conducted to determine N-telopeptide concentrations in bone samples.

The Significance of 3D Printed Firearms with Regards to Lethal Capacities and Traceable Elements
Caitlin Baker, B.A., Zara Ellen Wenzinger, M.S., Stephanie Wetzel, Ph.D., John Viator, Ph.D., Allison Laneve, M.S., Brian Kohlhepp, Pamela Marshall, Ph.D. - Duquesne University*

Metal detectors, gun-shot residue (GSR), serial numbers, and other commonly used firearm analysis methods have proven to be no match for 3D printed ghost guns in preliminary results. Three Liberator model guns were produced for this testing utilizing two polymers in the 3D printing process: acrylonitrile butadiene styrene (ABS) and polylactic acid (PLA). A remote trigger-device was used to fire the assembled weapons directly into a contained system to assess their lethality in humans. Gun remnants present at the conclusion of the firing process were collected for analysis including determination of structural integrity. This research advances knowledge and understanding of 3D printed firearm analysis.