MAAFS 2022 - Breakout Sessions - Schedule & Abstracts

Thursday, May 12

Scholarship Winners

1:30 pm **A Quantifiler™ Trio-based Mixture Screening Assay for the QuantStudio™ qPCR platform**
Dayanara Torres, B.S., Chastyn Smith, B.S., Andrea L. Williams, M.S., Edward L. Boone, Ph.D., Sarah Seashols-Williams Ph.D, and Tracey Dawson Green, Ph.D. - Virginia Commonwealth University*

2:05 pm **An Assessment of Probabilistic Approaches to mtDNA Mixture Interpretation**
Alyssa Addesso, Luigi Armogida, Brian Young, Jeffrey Smith, Mitchell Holland, Jennifer McElhoe - The Pennsylvania State University*

2:25 pm **In Vitro Metabolic Profile of a New Synthetic Opioid Bucinnazine Using Liquid Chromatography Tandem Mass Spectrometry (LC-MS/MS)**
Karissa N. Resnik, Emanuele A. Alves - Virginia Commonwealth University*

Interdisciplinary Session

2:45 pm **Not Just Another Acronym: FLN-TWG**
Linda Jackson - Virginia Department of Forensic Science

3:05 pm **ABC Update**
Danielle Hankinson - American Board of Criminalistics

3:20 pm **Break**

Biology Section - Pearl Ballroom

3:30 pm **SWGDAM Update Spring, 2022**
Lisa Schiermeier-Wood - Virginia Department of Forensic Science

4:00 pm **Optimization of a Microfluidic Device for Cell Capture of Spermatozoa Using Optical Trapping**
Samantha Pagel, Mackenzie Lally, M.S.; Ciara Rhodes, B.S.; Sarah J. Seashols-Williams, Ph.D., Dr. Joseph Reiner, and Dr. Tracey Dawson Green - Virginia Commonwealth University*

4:15 pm **Optimization and Comparison of Methods for Separation of Spermatozoa from Superabsorbent Polymer-Containing Forensic Evidence**
Hannah G. Wells, B.S.; Ciara Rhodes, B.S.; Sarah J. Seashols-Williams, Ph.D. - Virginia Commonwealth University*

4:30 pm **Reducing Interference in the UV-Vis Spectra of Blood Samples to Detect EDTA via the RED-BLEU Assay**
Kristen M. Atkinson, Brittany C. Hudson MSFS, Catherine Cupples Connon PhD - Virginia Commonwealth University*

Criminalistics Section - Grand Salon IV/V

3:30 pm **Real Time GCMS Analysis of Powders, Solids, Liquids, and more, using Agilent's QuickProbe™ Technology**
Kirk Lokits - Agilent Technologies

4:00 pm **The Analysis of Commercially Available Kratom Products in Richmond, VA**
James Fleming, Justin Poklis, Michelle Peace, Emanuele Alves - Virginia Commonwealth University*

4:15 pm **Forensic Pattern Analysis: A New Approach to Forensic Science Education**
Jonathan Fried - Loyola University Maryland

4:30 pm **Quantitative Analysis of Anticoagulants in Human Blood by Triple Quadrupole Mass Spectrometry**
Hiu Yu Lam, Tais Fiorentin, Francis Diamond, Amanda Mohr, Barry Logan - The Center for Forensic Science Research and Education*

4:45 pm **A Determination of the Aerosolization Efficiency of Drugs of Abuse in a Eutectic Mixture with Nicotine in Electronic Cigarettes**
Laerissa Reveil[*1], Adam C. Pearcy[2], Ph.D., Jazmine Povlick[3], M.S., Justin Poklis[4], B.S., Matthew S. Halquist[2], Ph.D., Michelle R. Peace[1], Ph.D. - [1]Department of Forensic Science, Virginia Commonwealth University; [2]Department of Pharmaceutics, Virginia Commonwealth University; [3]Department of General Services; [4]Department of Pharmacology & Toxicology, Virginia Commonwealth University*

Questioned Documents Section - Blue Point II

3:30 pm **The Use of Scientific Instruments to Forensically Examine Booklet Perforations**
George Virgin - HSI Forensic Laboratory

4:00 pm **Manual Reconstruction of Shredded Documents**
Meg OBrien - US Secret Service

MAAFS 2022 - Breakout Sessions - Schedule & Abstracts

Friday, May 13th

Biology Section - Pearl Ballroom

8:30 am **SWGDAM developmental validation section 3.2.6.1 Accuracy Studies for MaSTR™ Probabilistic Genotyping Software**
Sarah Copeland, Daniel Erb, Eric Podlaszewski, James Todd, Teresa Snyder-Leiby - Softgenetics*

8:45 am **Trace DNA: A Review**
Ashlie Roederer - Virginia Department of Forensic Science

9:15 am **Non-destructive method to estimate the number of contributors and predict DNA yield in touch DNA**
Christin Lee, Sarah Ingram, Christopher Ehrhardt, Susan Greenspoon - Virginia Commonwealth University / Virginia Department of Forensic Science*

9:45 am **Break**

10:00 am **Can I, Must I, Dare I Testify to a Reasonable Degree of Scientific Certainty?**
David H. Kaye - Yale Law School and Pennsylvania State University

10:20 am **Roundtable Discussion**
Moderator: Mimi Smith - Virginia Department of Forensic Science

11:00 am **Reproducibility of Quantifiler® Trio Assay-Specific Standard Curves Over a Range of Variables to Generate Guidelines for Crime Laboratory Workflows**
Stephanie M. Betts, MS, Kelly Knight, M.S.F.S[1], Joseph A. DiZinno, DDS[1], Megan M. Foley, M.S.F.S.[2] - [1]George Mason University, [2]The George Washington University*

11:15 am **Comparison of the MicroGEM forensicGEM Universal kit with the Qiagen QIAamp DNA**
Investigator kit for the extraction of DNA from saliva
Aubrey Shanahan, B.S. - Cedar Crest College

11:30 am **Determining Side of Deposition for Seminal Fluid Staining**
Jessica Kirby and Catherine Cupples Connon, PhD - Virginia Commonwealth University*

11:45 am **Destroying the Evidence: The Effects of Quicklime on Insect Colonization and Animal Scavenging**

Elle Winfield, Dr. Rebecca Forkner, Dr. Anthony Falsetti, Dr. Shawn T. Dash - George Mason University*

Criminalistics Section - Grand Salon IV/V

8:30 am **Premixed Fuel – A Unique Pattern**
Kaitlin E. Mertz, MSFS, ABC-FD - Virginia Department of Forensic Science

9:00 am **NIST Mass Spectral Libraries and Resources**
Edward Erisman - NIST

9:15 am **Application of a Validated QuEChERS extraction from Liver Tissue and analysis of 34 Fentanyl Analogs to Postmortem Liver**
Kylea Mathison, Sharon Kalb, Katherine Davis, Joseph Cox, Stephen Raso, Francisco Diaz, and Luis E. Arroyo - West Virginia University*

9:30 am **Forensic Science Research and Development: An Overview of Grant Funded Outcomes**
Jennifer Love and Tracey Johnson - National Institute of Justice*

10:00 am **Break**

10:15 am **Feasibility study for on-site screening of organic and inorganic gunshot residue using portable electrochemical devices**
Kourtney Dalzell, Colby E. Ott, M.S., Tatiana Trejos, Ph.D., Luis E. Arroyo, Ph.D. - West Virginia University*

10:30 am **Control Cans for Fire Debris and Why They are Needed**
Michelle M. Drake, M.S. - Virginia Department of Forensic Science

11:00 am **Sampling Uncertainty for the Extrapolation of Calculated Collective Net Weights**
Christopher Merrill - Allegheny County Medical Examiner's Office

11:15 am **HPLC/LCMS-MS Analysis of Heroin, Fentanyl, Fentanyl Analogs, and Common Adulterants**
Ryki B. Vance, B.S, MPS[*1]; Hariny M. Isoda, B.S, MPS[1], William H. Campbell, PhD[1]; Mandy L. Tinkey[2]; Matthew R. Wood, PhD[3]*
[1]Penn State University, University Park, PA; [2]Allegheny County Medical Examiner's Office, Pittsburg, PA; [3]Ocean County Sheriff's Office, Ocean County, NJ

11:45 am **Evaluation of the analytical performance of a portable 1064 nm Raman instrument on simulated drug mixtures and authentic case samples using deep learning**
Alexis N. Wilcox[*1], Colby E. Ott[1], Travon Cooman[1], Amber Burns[3], Edward Sisco[2], Luis E. Arroyo[1]*
[1]West Virginia University, [2]National Institute of Standards and Technology (NIST), [3]Maryland State Police Forensic Laboratory

Questioned Documents Section - Blue Point II

There are currently no Questioned Documents presentations scheduled for Friday due to a lack of submissions. The Biology and Criminalistics section presentations are open to all meeting attendees.

Interdisciplinary Session Abstracts

Not Just Another Acronym: FLN-TWG

Linda Jackson - Virginia Department of Forensic Science

In 2018, the National Institute of Justice created the Forensic Laboratory Needs Technology Working Group (FLN-TWG) to improve federal coordination with state and local forensic science laboratories. The main goal of the FLN-TWG was to inform NIJ's decision-making process on research and implementation of forensic technology to ensure that they are relevant and responsive to the laboratory operations needs of the forensic science community.

The 24-member group includes laboratory directors from across the country, representing a diversity of jurisdictions and laboratories. Representatives from independent laboratories and those organized by law enforcement agencies are included, as well as leaders in the field of forensic science research. The working group is supported by the Forensic Technology Center of Excellence.

FLN-TWG has published four white papers that highlight implementation considerations and strategies for developing methodologies:

- LC-MS-Based Forensic Toxicology Screening
- Proteomic Mass Spectrometry for Biology Fluid Identification
- Next Generation Sequencing for DNA Analysis
- 3D Imaging for Firearms and Toolmarks

In 2021, the FLN-TWG divided into subcommittees to begin working on additional topics. The goals and deliverables of each subcommittee, including Research, LIMS, Technology Needs, and Novel Seized Drug Technologies, will be described.

Further information and white papers can be accessed on the FLN-TWG website: https://forensiccoe.org/forensic-laboratory-needs-technology-working-group/.

ABC Update

Danielle Hankinson - American Board of Criminalistics

ABC update to include organizational updates on accreditation, examinations, recertification, etc.

Biology Section Abstracts

A Quantifiler™ Trio-based Mixture Screening Assay for the QuantStudio™ qPCR platform
Dayanara Torres, B.S., Chastyn Smith, B.S., Andrea L. Williams, M.S., Edward L. Boone, Ph.D., Sarah Seashols-Williams Ph.D, and Tracey Dawson Green, Ph.D. - Virginia Commonwealth University*

At present, the forensic DNA workflow is not capable of providing information about the contributor status (single vs multiple contributors) of evidentiary samples prior to end-point analysis. Due to this shortcoming, a high-resolution melt (HRM) curve assay was previously developed and integrated into the Investigator Quantiplex® qPCR kit and optimized for use on the QuantStudio™ 6 qPCR platform. When tested, this assay was able to accurately distinguish between single source and mixture samples 87.88% of the time. As a result of this success, integration of the HRM assay into a more commonly used chemistry, the Quantifiler™ Trio kit, was pursued. Integration of the HRM assay into the Quantifiler™ Trio required redesign of the HRM assay, encompassing inclusion and calibration of a new intercalating dye, optimization of reaction conditions, and an adjustment of data analysis settings. Once these modifications were incorporated into a formal final protocol, the assay was able to accurately identify 73.8% of all single source and mixture samples tested.

An Assessment of Probabilistic Approaches to mtDNA Mixture Interpretation
Alyssa Addesso, Luigi Armogida, Brian Young, Jeffrey Smith, Mitchell Holland, Jennifer McElhoe - The Pennsylvania State University*

An interface between two previously developed software tools, Mixemt and MixtureAce™, is being evaluated as a tool for deconvoluting mitochondrial DNA mixtures; MixtureAce™ is utilized for filtering noise while Mixemt is utilized to determine the haplogroups and haplotypes. To evaluate the accuracy of the software to correctly identify the haplogroups and proportions of the individuals comprising the mixture, both in-silico and biological datasets were used. In-silico mixtures served to test and evaluate the MixtureAce™-Mixemt pipeline, while biological mixtures served to further assess the software tools, ensuring the applicability of the software to forensic casework and accounting for unpredictable laboratory variables. Mixtures comprised various combinations of mtDNA haplotypes, from closely related to vastly different SNP/INDEL profiles. This work could impact the ability and practices of the forensic science community to deconvolute mtDNA mixtures and serve as additional motivation for a broader range of laboratories to adopt MPS mixed mtDNA analysis.

SWGDAM Update Spring, 2022
Lisa Schiermeier-Wood - Virginia Department of Forensic Science

The Scientific Working Group on DNA Analysis Methods, known as SWGDAM, is currently comprised of dedicated forensic scientists from international, federal, state and local forensic DNA laboratories as well as guests representing academia and other Federal agencies who offer their expertise in DNA technologies. SWGDAM serves as a forum to discuss, share, and evaluate forensic biology methods, protocols, training, and research to enhance forensic biology services as well as provide recommendations to the FBI Director on quality assurance standards for forensic DNA analysis.
Updates on SWGDAM's standing committees will be presented in addition to information on current topics being addressed by SWGDAM working groups.

Optimization of a Microfluidic Device for Cell Capture of Spermatozoa Using Optical Trapping
Samantha Pagel, Mackenzie Lally, M.S.; Ciara Rhodes, B.S.; Sarah J. Seashols-Williams, Ph.D., Dr. Joseph Reiner, and Dr. Tracey Dawson Green - Virginia Commonwealth University*

Optical trapping is a novel front-end separation technique that can be used to manipulate dielectric particles, such as cells. In heterogeneous mixtures, optical tweezers can be used to manipulate and isolate cells of interest from other cell types. For this research, a range of 5 to 45 sperm cells were isolated from 1:20 diluted neat semen and an equal volume mixture of 1:20 semen mixed with resuspended vaginal epithelial cells. The collected sperm cells were extracted and carried through the entire DNA analysis process ending with profile interpretation. Partial and full profiles of the male donor were obtained with an average trapping time of 2 minutes per cell and an average of 1 allele or less that could be attributed to the female contributor observed in the profile. Optical trapping is a promising alternative technique for isolating forensically relevant cells of interest, such as spermatozoa, from other cell types in heterogeneous mixtures.

Optimization and Comparison of Methods for Separation of Spermatozoa from Superabsorbent Polymer-Containing Forensic Evidence
Hannah G. Wells, B.S.; Ciara Rhodes, B.S.; Sarah J. Seashols-Williams, Ph.D. - Virginia Commonwealth University*

Sexual assault is currently one of the most prevalent crimes that affects victims of all ages. Forensic DNA analysts often confirm proof of contact in sexual assault cases through the identification of spermatozoa and subsequent STR profiling. Diapers and other feminine hygiene products, such as maxi pads and ultrathin pads, are types of superabsorbent polymer-containing evidence that complicate the process of DNA analysis due to the trapping of the spermatozoa in the gel-like matrix. In this study, a comparison of methods was performed to determine which extraction technique produces the highest sperm cell and DNA yield and generates a usable STR profile. When comparing the previously reported centrifugal filtration and calcium chloride ($CaCl_2$) dehydration methods, a teasing and filtration method, and a sodium chloride (NaCl) and filtration method, microscopic examination demonstrated statistically significant sperm yields using the sodium chloride method for the diaper samples and the teasing and filtration method for the remaining substrate types. However, when quantifying the DNA extracts obtained from the substrates, most methods resulted in similar DNA concentrations, with little indication of degradation or inhibition. All four extraction methods produced full STR profiles with no indication of inhibition or degradation. This suggests that forensic laboratories have the flexibility to choose among these spermatozoa extraction methods when analyzing superabsorbent polymer-containing evidence.

Reducing Interference in the UV-Vis Spectra of Blood Samples to Detect EDTA via the RED-BLEU Assay
Kristen M. Atkinson, Brittany C. Hudson MSFS, Catherine Cupples Connon PhD - Virginia Commonwealth University*

A rapid method to support or refute claims of planted blood evidence in the courtroom can lead to increased transparency in criminal cases and ensure justice. Thus, we have continued to optimize the RED-BLEU assay, which is a colorimetric test followed by ultraviolet-visible spectroscopy to presumptively detect the presence of a common anticoagulant, EDTA. While the use of the NanoDrop™ 2000 spectrophotometer reduced subjectivity and enabled quantification of EDTA within samples, other components from blood exhibited substantial interference with this portion of the assay. In this study, efforts were made to reduce this interference using several treatment methods known to precipitate or

denature proteins. Ammonium sulfate, trichloroacetic acid, sodium hydroxide, trypsin, and proteinase K were evaluated, with proteinase K demonstrating the most promise for removing the blood protein interference when implemented between the colorimetric test and UV-visible spectroscopy.

SWGDAM developmental validation section 3.2.6.1 Accuracy Studies for MaSTR™ Probabilistic Genotyping Software

Sarah Copeland, Daniel Erb, Eric Podlaszewski, James Todd, Teresa Snyder-Leiby - Softgenetics*

SWGDAM developmental validation guidelines include: “3.2.6 Accuracy…. - 3.2.6.1 These studies should include the comparison of the results produced by the probabilistic genotyping software to manual calculations, or results produced with an alternate software program or application, to aid in assessing accuracy of results generated by the probabilistic genotyping system. Calculations of some profiles (e.g., complex mixtures), however, may not be replicable outside of the probabilistic genotyping system”

Manual calculations were performed in Excel and in MaSTR software to evaluate accuracy of implementation of HWE, inbreeding and co-ancestry following NRCII Recommendations 4.1 and 4.2, using a single source sample allele calls and RFUs. Manual calculations were performed to evaluate accuracy of Likelihood Ratio calculations from weighted genotypes obtained for two-person mixture analysis in MaSTR software. More complex mixtures of 2, 3 and 4 contributors were analyzed in MaSTR software and EuroForMix. The results between the two PG software programs are concordant.

Trace DNA: A Review

Ashlie Roederer - Virginia Department of Forensic Science

A review of trace DNA in the literature that characterizes success rates of trace DNA in casework samples, categorized by type of sample and type of offense, the likelihood of getting a usable profile for trace DNA submissions. This lecture will also discuss why trace DNA is hard to find and collect, and how the nature of DNA affects the success rates.

Non-destructive method to estimate the number of contributors and predict DNA yield in touch DNA

Christin Lee, Sarah Ingram, Christopher Ehrhardt, Susan Greenspoon - Virginia Commonwealth University / Virginia Department of Forensic Science*

Touch or trace DNA samples are frequently difficult to interpret and determine the number of contributors, and these challenges can decrease the probative value of these samples. Therefore, we developed a non-destructive molecular technique to resolve the mixture status of a touch/trace sample and predict DNA yield using fluorescent-tagged antibodies to target testosterone, dihydrotestosterone, and cytokeratins in epithelial cells. The results demonstrated that hormones were able to distinguish each contributor and be used as an individualizing signature for estimating the number of contributors in touch/trace DNA mixtures. Also, the data showed strong correlations between DNA quantity and the probe binding efficiency, indicating this method can predict DNA yield before DNA profiling. These findings indicate that antibody hybridization has the potential to serve as a non-destructive technique prior to STR analysis to enhance the probative value of biological evidence.

Can I, Must I, Dare I Testify to a Reasonable Degree of Scientific Certainty?
David H. Kaye - Yale Law School and Pennsylvania State University

Forensic-science experts may be asked or expected to testify to "a reasonable degree of scientific certainty," a "reasonable degree of ballistic certainty," or a "reasonable degree" of some other discipline-specific "certainty." Is this magic phrase legally necessary? What does it mean in the eyes of the law? What can an expert who is reluctant to use it do? This talk will offer some answers and suggestions.

Reproducibility of Quantifiler® Trio Assay-Specific Standard Curves Over a Range of Variables to Generate Guidelines for Crime Laboratory Workflows
Stephanie M. Betts, MS, Kelly Knight, M.S.F.S[1], Joseph A. DiZinno, DDS[1], Megan M. Foley, M.S.F.S.[2] - [1]George Mason University, [2]The George Washington University*

Quantitative PCR (qPCR) is the preferred method of quantitation in forensic DNA analysis to determine the amount of amplifiable human DNA present using a standard curve containing known DNA concentrations. The primary goal of this research was to generate laboratory guidelines and recommendations for quantitation standards for forensic laboratories hoping to streamline their workflows, and to determine how long standards are valid to decrease the amount of time and money spent on assay-specific standard curves. The research data was generated over a two-month period, in which Quantifiler® Trio manufacturer guidelines were followed for the creation of five concentrations varying from 50 ng/µL to 0.005 ng/µL and analyze the following variables; variation in curve parameters, including inter-run variability and reproducibility between analysts, and efficacy of the assay-specific standards versus a virtual curve using mock casework samples. Results indicated little to no difference in the values for T.Y, small, and large autosomal targets when both analysts' curve parameter values were compared, linearity remained consistent beyond the recommended discard of 14-days, and the data also showed little to no difference between the two curve methods prior to amplification, assay-specific standard curves, or virtual standard curves. Additionally, three positive control values, 5, 1 and 0.5 ng/uL, were evaluated with consistency up to 42-days.

Comparison of the MicroGEM forensicGEM Universal kit with the Qiagen QIAamp DNA Investigator kit for the extraction of DNA from saliva
Aubrey Shanahan, B.S. - Cedar Crest College

While there are many commercially available DNA extraction kits, few use a procedure as fast and as simple as the MicroGEM forensicGEM Universal kit. Although it is a newer kit, recent research has shown this Temperature Driven Extraction method to be capable of extracting more DNA from touch DNA samples than the Qiagen QIAamp DNA Investigator kit. This research aims to show how the forensicGEM Universal kit compares to the Qiagen QIAamp DNA Investigator kit in the processing of saliva samples. Using six substrates and three types of swabs, various combinations will be explored to determine the overall percent recovery of saliva DNA in both extraction kits.

Determining Side of Deposition for Seminal Fluid Staining
Jessica Kirby and Catherine Cupples Connon, PhD - Virginia Commonwealth University*

The purpose of this study was to assess if there is an objective way to determine which side of a substrate a seminal fluid stain was deposited. Two volumes (100ul and 250ul) of fresh semen were deposited in triplicate onto three different substrates (denim, cotton, and polyester). Measurements were taken of the resulting stains on both sides of the fabric, followed by swabbing both sides of each stain for testing using Acid Phosphatase (AP) and Kernechtrot-Picroindigocarmine Staining (KPICS), as well as DNA extraction, quantification, and STR profile development. Results were evaluated to determine if there was a significant difference in stain size, detection of acid phosphatase, sperm count, DNA quantitation, and/or STR profiles between the two sides of the substrate, with the overall goal of using such information to predict side of deposition.

Destroying the Evidence: The Effects of Quicklime on Insect Colonization and Animal Scavenging
Elle Winfield, Dr. Rebecca Forkner, Dr. Anthony Falsetti, Dr. Shawn T. Dash - George Mason University*

Insect activity and scavenger visitation on surface deposited remains often vary by location but will also vary by condition of the remains. After a homicide, perpetrators often alter evidence by burning remains or covering remains with chemical substances such as quicklime. In cases involving outdoor remains disposal, all three of these factors – insects, scavengers, and treatment of remains – may interact to alter rates of tissue loss. To determine the individual effects of different body conditions, I assessed insect colonization and scavenger visitation events of quicklime covered and charred pig ribs in a forested region in Northern Virginia. In three forested blocks within the George Mason University Forensic Science and Research Training Lab facility, I placed three replicates each of three treatments (1) charred, (2) lime covered, (3) untreated (control) pig ribs. In addition to general surveys of insect colonization and vertebrate scavenging events, I used wildlife cameras to monitor vertebrate visitation. Results demonstrated that neither insects nor vertebrates were deterred by charring or lime. Adult fly visitation was significantly higher on lime-treated remains. Insect colonization was highest eight days 8 after deposition, evident by maggot masses on the undersides of at least 3 ribs per block. I encountered several medium-sized vertebrate scavengers, including red fox (Vulpes vulpes), possum (Didelphis virginiana), and raccoon (Procyon lotor). Although there were block-level differences in the number of scavenger visits, burning and lime treatment did not deter scavengers. This study suggests that investigators who are specifically dealing with charred or limed remains may still need to account for insect removal of tissue and vertebrate scavenging.

Criminalistics Section Abstracts

In Vitro Metabolic Profile of a New Synthetic Opioid Bucinnazine Using Liquid Chromatography Tandem Mass Spectrometry (LC-MS/MS)

Karissa N. Resnik, Emanuele A. Alves - Virginia Commonwealth University*

Bucinnazine, also known as AP-237, is a synthetic opioid recently discovered in seized heroin samples in the U.S. and in Europe, though at present, bucinnazine is not scheduled in the U.S., as it is not a therapeutic choice for the treatment of pain. Nevertheless, with the advent of the crypto currency and the easy access of substances on the Darknet, bucinnazine is a real threat to the public health. The metabolic profile is an important tool for the development of analytical methods for the screening of emerging synthetic opioids, as the metabolites act as biomarkers of previous use. The aim of this study is to identify the in vitro phase I/II metabolites of bucinnazine by using pooled rat liver microsomes. Given the increase in illicit bucinnazine cases in the United States, a deeper understanding of its metabolites as biomarkers is important for the development of reliable identification strategies to detect bucinnazine misuse cases.

Real Time GCMS Analysis of Powders, Solids, Liquids, and more, using Agilent's QuickProbe™ Technology

Kirk Lokits - Agilent Technologies

This presentation demonstrates the capabilities of performing fast GCMS analysis in under 1 minute, requiring minimal to no sample preparation prior to analysis, and utilizes classical EI commercial libraries. This work seeks to illustrate how Agilent's QuickProbe™ can be used as a fast-qualitative screening tool on an existing 5977B/7890B or 8890 GCs, while allowing for the continued use of a co-resident split/splitless GC inlet for routine conventional capillary GCMS analysis.

The Analysis of Commercially Available Kratom Products in Richmond, VA

James Fleming, Justin Poklis, Michelle Peace, Emanuele Alves - Virginia Commonwealth University*

Kratom is a novel psychoactive substance that has gained popularity within the past ten years due to its activity at the μ-opioid receptor. Twenty-nine kratom samples were obtained from tobacco and vape shops in the Richmond, Virginia area, including powders, teas, capsules, extracts, and a carbonated beverage. A qualitative, organic profile was obtained with Direct Analysis in Real Time-Mass Spectrometry (DART-MS) and confirmed with Gas Chromatography-Mass Spectrometry (GC-MS). A quantitative inorganic profile was obtained with Inductively Coupled Plasma-Optical Emission Spectroscopy (ICP-OES). As a herbal product, no quality control is expected from manufacturers, which brings a public health concern due to the association of high concentrations of active substances and toxic metal levels in commercial kratom samples.

Forensic Pattern Analysis: A New Approach to Forensic Science Education
Jonathan Fried - Loyola University Maryland

The vast majority of forensic science programs provide training and coursework focused broadly on criminalistics and specifically on either crime scene investigation or forensic biology and chemistry. Prior to this academic year, no undergraduate or graduate programs existed in Maryland or the mid-Atlantic region that focused primarily on the comparative sciences. To overcome this perceived deficiency, Loyola University Maryland has developed a Master of Science degree in Forensic Pattern Analysis. During the course of curriculum design, numerous forensic science practitioners at state, local and federal agencies were consulted on the needs for current and future scientists engaged in latent prints casework. This presentation will discuss the feedback received and attempt to place the identified needs and recommendations into the context of a graduate curriculum focused on forensic pattern evidence analyses.

Quantitative Analysis of Anticoagulants in Human Blood by Triple Quadrupole Mass Spectrometry
Hiu Yu Lam, Tais Fiorentin, Francis Diamond, Amanda Mohr, Barry Logan - The Center for Forensic Science Research and Education*

Two major anticoagulant outbreaks occurred in Chicago in 2018 and Tampa in 2021, where synthesis cannabinoids were laced with anticoagulant drugs. Both incidents resulted in at least 5 deaths and more than 200 individuals hospitalized. It is challenging for traditional drug checking workflow to detect these compounds due to small amount present in sample and their chemical properties. A unique and comprehensive analytical assay was proposed for analysis of anticoagulants in human blood.

A Determination of the Aerosolization Efficiency of Drugs of Abuse in a Eutectic Mixture with Nicotine in Electronic Cigarettes
Laerissa Reveil[*1], Adam C. Pearcy[2], Ph.D., Jazmine Povlick[3], M.S., Justin Poklis[4], B.S., Matthew S. Halquist[2], Ph.D., Michelle R. Peace[1], Ph.D. - [1]Department of Forensic Science, Virginia Commonwealth University; [2]Department of Pharmaceutics, Virginia Commonwealth University; [3]Department of General Services; [4]Department of Pharmacology & Toxicology, Virginia Commonwealth University*

The adulteration of e-liquids with drugs other than nicotine can form a eutectic mixture, lowering the melting point to promote aerosolization of drugs. This study evaluated the aerosolization and recovery of nicotine and methadone in an electronic cigarette using a validated Cooperation Centre for the Scientific Research Relative to Tobacco (CORESTA) method in an automated vaping machine with the following parameters: an inhale duration of 3 seconds, an exhale duration of 10 seconds, and a puff volume of 60 mL for a total of 15 puffs. Aerosols from four e-liquids (pure nicotine and methadone hydrochloride at 12 mg/mL in 1:1 PG:VG, 1:1 methadone hydrochloride:nicotine at 12 mg/mL in 1:1 PG:VG, and 1:1 PG:VG) were captured and analyzed using a Gas Chromatograph-Mass Spectrometer (GC-MS) to determine the amount of drugs aerosolized. In the single drug e-liquids (n = 3), 1.6 ± 0.2 mg of nicotine was aerosolized while 2.7 ± 0.1 mg of methadone was aerosolized, and in the mixed drug e-liquid (n = 3), both nicotine and methadone increased to 3.7 ± 0.5 mg and 3.7 ± 0.1 mg, respectively. E-liquids containing nicotine and methadone created a eutectic mixture that increased the amount of drug delivered in the aerosol.

Premixed Fuel – A Unique Pattern
Kaitlin E. Mertz, MSFS, ABC-FD - Virginia Department of Forensic Science

Fire Debris analysis involves the examination of materials collected at a fire scene for the presence of ignitable liquids. The data produced by the analysis of ignitable liquids generates patterns that allow for their classification. In the fall of 2019, the Virginia Department of Forensic Science analyzed a liquid and a sample of fire debris for the presence of an ignitable liquid. The data for each sample contained a unique pattern not previously seen. This presentation will outline the steps taken to identify the pattern, to determine the type of product it could have originated from, and additional information about premixed fuels that has been learned since.

NIST Mass Spectral Libraries and Resources
Edward Erisman - NIST

The NIST Mass Spectrometry Data Center maintains the NIST mass spectral libraries and provides free mass spectrometry software. This talk will go over the tools available and information about updates to the libraries. The 2020 release of the EI mass spectral library contains spectra for 306,643 compounds and retention index values for 139,382 compounds. The 2020 release of the tandem library contains 1.3 million spectra from 31,000 compounds. NIST provides software for EI mass spectral extraction, searching, and interpretation.

Application of a Validated QuEChERS extraction from Liver Tissue and analysis of 34 Fentanyl Analogs to Postmortem Liver
Kylea Mathison, Sharon Kalb, Katherine Davis, Joseph Cox, Stephen Raso, Francisco Diaz, and Luis E. Arroyo - West Virginia University*

Postmortem liver tissue from 16 deidentified sources was surveyed, targeting a suite of 34 fentanyl analogs including 8 deuterated internal standards using a validated QuEChERS (Quick Easy Cheap Effective Rugged and Safe) extraction protocol coupled to a liquid chromatography-tandem mass spectrometry (LC-MS/MS) method. Liver samples were homogenized with a high-speed mixer mill with house-made attachments to facilitate the pulverization of the sample before extraction. The QuEChERS extraction was modified to accommodate smaller specimen quantities (0.1 g of liver tissue) and reduced costs by using disposable materials. The procedure utilized magnesium sulfate, sodium chloride, 3 stainless steel balls, and 2 mL dispersive-SPE tubes containing 25 mg of primary secondary amine and 25 mg end-capped octadecylsilane (C18EC). All the liver specimens were positive for fentanyl, ranging from 6 µg/kg to >100 µg/kg, with an average concentration of 92 µg/kg. The most frequently found metabolites were 4-ANPP at an average of 9 µg/kg and norfentanyl at an average of 20 µg/kg. In 7 of the 16 (44 %) samples analyzed, more than one fentanyl analog was detected, and the three most common fentanyl analogs detected were methoxyacetyl fentanyl, acetyl fentanyl and alpha-methyl acetyl fentanyl.

Forensic Science Research and Development: An Overview of Grant Funded Outcomes
Jennifer Love and Tracey Johnson - National Institute of Justice*

National Institute of Justice (NIJ) Office of Investigative and Forensic Sciences (OIFS) is the lead federal agency for forensic science research and development and manages programs and projects that inform the federal, state, and local forensic science communities. Its mission is to improve the quality and practice of forensic sciences through innovative solutions that support research and development, testing and evaluation, technology, and information exchange for the criminal justice community. NIJ has recently funded several exciting research projects through two grant programs: Research and Evaluation for the Testing and Interpretation of Physical Evidence in Publicly Funded Forensic Laboratories and Research and Development in Forensic Science for Criminal Justice Purposes. An overview of some of the project outcomes funded through these grants will be presented.

Feasibility study for on-site screening of organic and inorganic gunshot residue using portable electrochemical devices
Kourtney Dalzell, Colby E. Ott, M.S., Tatiana Trejos, Ph.D., Luis E. Arroyo, Ph.D. - West Virginia University*

Reliable laboratory and onsite screening testing for rapid decision-making and investigative leads is still a pending task in the field of gunshot residues (GSR). Efficient identification of inorganic and organic gunshot residue demands faster, more specific, and more sensitive methods. Electrochemistry demonstrates the ability to detect inorganic (IGSR) and organic gunshot residues (OGSR) simultaneously using a small volume, low-cost, disposable screen-printed carbon electrode platform. In this study, benchtop and portable electrochemical systems were compared for performance on a panel of IGSR and OGSR analytes consisting of 7 common explosives, stabilizers, and plasticizers. The two instruments were tested on a population of authentic hand residues comprising 200 non-shooters, 100 leaded-ammunition shooters, and 50 lead-free ammunition shooter samples. Performance measures including false positive and false negative rates were calculated, and an accuracy of 95.7% and 96.5% for the benchtop and portable instruments was achieved, respectively. The outcomes of this work demonstrate not only the speed and convenience of simultaneous IGSR and OGSR detection but also the effectiveness of the portable technology, showcasing the future of field portable GSR detection and potential breakthrough for more effective case management in firearm-related investigations.

Control Cans for Fire Debris and Why They are Needed
Michelle M. Drake, M.S. - Virginia Department of Forensic Science

Fire debris samples for ignitable liquid residue analysis are frequently packaged in metal paint cans. Sealed paint cans are considered airtight, thereby maintaining the integrity of the evidence and preventing evaporation of any ignitable liquids that may be present. Changes in manufacturing processes of paint cans have resulted in light to medium aromatic products being present in some new, unused gray epoxy lined paint cans, which could directly interfere with the analysis of items packaged within those cans. This presentation demonstrates the value of submitting a control can along with evidence samples in all cases and how it strengthens the case results.

Sampling Uncertainty for the Extrapolation of Calculated Collective Net Weights
Christopher Merrill - Allegheny County Medical Examiner's Office

Sampling uncertainty must be accounted for when extrapolating a calculated collective net weight. Statistical calculations can be performed to account for this while assessing variables (including net powder weights versus indirect weighing, weighing events, expansion values (k) for statistical calculations, and rounding versus truncating values) to determine the best practice for samples seized within Allegheny County. A calculator was designed to perform these calculations given the variables with further input from members at the Drug Enforcement Agency and Pennsylvania State Police to provide a more accurate representation for reporting uncertainty of these calculated weights.

HPLC/LCMS-MS Analysis of Heroin, Fentanyl, Fentanyl Analogs, and Common Adulterants
Ryki B. Vance, B.S, MPS[*1]; Hariny M. Isoda, B.S, MPS[1], William H. Campbell, PhD[1]; Mandy L. Tinkey[2]; Matthew R. Wood, PhD[3]*
[1]Penn State University, University Park, PA; [2]Allegheny County Medical Examiner's Office, Pittsburg, PA; [3]Ocean County Sheriff's Office, Ocean County, NJ

The goal of this study was to achieve an optimized method of separation utilizing high-pressure liquid chromatography-tandem mass spectrometry (HPLC/ Tandem LCMS) that is simple, quick, retains baseline resolution, and uses materials that crime labs commonly utilize. LCMS is emerging as a powerful tool to be used adjacently with and/or potentially replace GCMS in many laboratories, and method development for LCMS is necessary for the advancement of this technology in the crime lab. Starting with HPLC analysis, once a satisfactory HPLC method was created it was then transferred to LCMS/MS for testing. The compounds were studied individually through infusions to identify optimal s-lens values, collision energies, and product ions which were used to optimize the method created and then subsequently applied to a mixture to access the quality of separation. Lastly, statistical analysis was conducted to validate the results obtained prior to implementing the method to evaluate seized drug samples.

Evaluation of the analytical performance of a portable 1064 nm Raman instrument on simulated drug mixtures and authentic case samples using deep learning
Alexis N. Wilcox[*1], Colby E. Ott[1], Travon Cooman[1], Amber Burns[3] , Edward Sisco[2] , Luis E. Arroyo[1]*
[1]West Virginia University, [2]National Institute of Standards and Technology (NIST), [3]Maryland State Police Forensic Laboratory

Raman spectroscopy plays a crucial role in drug identification as a rapid, non-destructive screening technique using portable instruments in the field, without opening packages and increasing safety of the operators. However, a major disadvantage of Raman spectroscopy is the limitation to identify drug analytes and diluents/cutting agents when present in mixtures. We investigated the analysis of simple mixtures and authentic case samples using convolutional neural networks (CNN)— a deep learning method able to select important features in a spectrum and classify compounds with high accuracy. Ninety-nine percent of all mixtures had at least one analyte correctly identified which demonstrated the usefulness of CNNs to improve the identification of compounds in mixtures as a screening technique.

Questioned Documents Section Abstracts

The Use of Scientific Instruments to Forensically Examine Booklet Perforations
George Virgin - HSI Forensic Laboratory

Many travel documents are in booklet format, with multiple security features to include perforations. Perforations have proven to be an effective device for securing the cover and interior pages of the booklet, to guard against the removal and substitution of pages. Forgers have become highly skilled in counterfeiting and tampering with booklet perforations, as a means to perfect the substitution of pages in travel documents. The deception is compounded in cases of fraudulent travel documents with booklet perforations that have been altered or counterfeited.

Forensic document examiners are often the last line of defense in detecting document fraud. The forensic examination of perforations can be an important aspect in detecting forged travel documents. Advancements in scientific instrumentation, and the incorporation of advanced examination techniques in casework, allow forensic document examiners to stay ahead of the forgers.

After attending this presentation, attendees will have learned about a preliminary study where scientific instrumentation at the Forensic Laboratory has been applied to examine booklet perforations in genuine and forged travel documents.

Manual Reconstruction of Shredded Documents
Meg OBrien - US Secret Service

Presentation on a case that required manual sorting and reconstruction of cross-cut shredded documents.