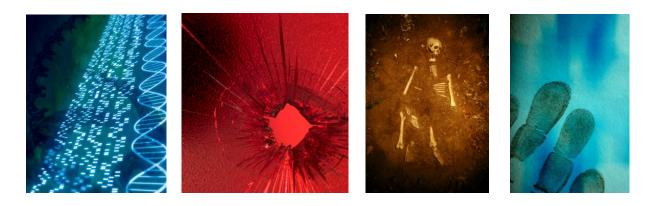


# Forensic Science & Law Graduate Research Symposium



## April 11-12, 2019 Fisher Hall Room 719





## Graduate Research Symposium Day 1

Thursday, April 11, 2019 Fisher Hall Room 719

8:00am     What Can You Learn from a Bag of Bones: The Utilization of Osteological and DNA Analysis of Human Skeletal Remains from the Flevaeis Plot Archaeological     McKenna Lohr       8:30am     Utilizing DNA Genotyping and Next Generation Sequencing in Identifying Bacteria     Sarah Lundholm and Creating Microbiomes Unique to Individuals       9:00am     The Use of DNA Analysis and Bacterial Profiles to Link Cats to an Institutionalized Hoarding Facility     Diana Delgadillo       9:30am     BREAK     9:45am     Optimization of an LC-MS/MS Method for the Separation and Identification of Synthetic Cannabinoids on Prison Mail Samples     Alyssa Hurd       10:15am     The Detection of Curcumin in Turmeric Dietary Supplements via LC/MS-MS     Kaitlin King       10:45am     Perception of Dominance of Individuals based on the Variability of Context     Katherine Wodyka       11:15pm     LUNCH     LUNCH     Lindsay Loughner       12:30pm     Genetic variation in the human relaxin 2 ( <i>RLN2</i> ) promoter affects <i>in vitro</i> Lindsay Loughner       expression, with potential implications for susceptibility to preterm birth     Lindsay Loughner       1:30pm     BREAK     Evaluation of Reference Profiles Created Using the Kinship Feature of     Erin Monko       2:15pm     Synthesis of Lanthanide-Doped Gahnite for Use in Fluorescent Detection of Gun- Joshua Kotchey     Joshua Kotchey </th <th>Time</th> <th>Title</th> <th>Presenter</th>	Time	Title	Presenter
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4:30pm Using Cell-Free DNA to Improve STR Analysis of Sweat Samples Nathan McFadden	4:00pm		Shannon Mahoney
	4:30pm	Using Cell-Free DNA to Improve STR Analysis of Sweat Samples	Nathan McFadden

#### 8:00am McKenna Lohr

### What Can You Learn from a Bag of Bones: The Utilization of Osteological and DNA Analysis of Human Skeletal Remains from the Flevaeis Plot Archaeological Site in Rhodes

Osteological and DNA analysis can be utilized on human skeletal remains found at a crime scene or archaeological site to distinguish between individuals and determine identifying characteristics. In the case of the Flevaeis Plot archaeological site in Rhodes, two graves were uncovered each containing six to seven skeletons. By utilizing the contextual information of the site, the bones are estimated to be 1,368 to 2,341 years old. Osteological analysis was performed on the remains brought to Duquesne University to determine size/intactness of each bone, the type of bone, side of origin, and in some cases sex, age, and overall health. Based on this data, it is estimated there are twelve bones total from at least four individuals. DNA analysis of hypervariable region I (HVI) within the mitochondrial DNA (mtDNA) genome was performed to differentiate between skeletal samples of divergent maternal lineage and construct haplogroups to determine maternal lineage. HVI sequences of the skeletal remains were compared to HVI sequences from studies of contemporary populations that reflect the ethnic groups that occupied Rhodes during the estimate time range in order to infer ethnic group of origin. (*Advisor - Lisa Ludvico*) (*Evaluator - Philip Reeder*) (*External Reviewer - Anne Burrows*)

8:30am Sarah Lundholm

#### Utilizing DNA Genotyping and Next Generation Sequencing in Identifying Bacteria and Creating Microbiomes Unique to Individuals

Microbiomes are communities of microorganisms including bacteria, archaea, protozoans and arthropods that inhabit a specific environment. Recent research has found that microbiomes found on and throughout the human body are unique to individuals, including identical twins, and thus have the potential to be used as an identification technique. The resistance and abundance of these microbes is advantageous in forensic analysis, particularly in cases of low copy number DNA. In this study, the microbiomes of 15 volunteers were analyzed. Participants handled a sterilized object and also provided swabs of their palms and mouth. A modified silica DNA extraction method was utilized followed by amplification via PCR. Amplified samples were analyzed with gel electrophoresis or Next Generation Sequencing. Time permitting, samples will also be run with capillary electrophoresis. The results are compared to determine what technique is optimal for integrating microbiomes into the forensic science field.

(Advisor – Becky Morrow) (Evaluator - Lisa Ludvico) (External Reviewer– Nancy Trun)

9:00am Diana Delgadillo

#### The Use of DNA Analysis and Bacterial Profiles to Link Cats to an Institutionalized Hoarding Facility

Animal cruelty is a serious crime that can reach the level of felony charges nationwide. Animal hoarding is a specific type of animal cruelty associated with unsanitary living conditions, lack of veterinary care, and a multitude of infectious diseases. The purpose of this study was to identify the bacterial profile in cats from an institutionalized hoarding facility (IHF) compared to non-IHF cats through DNA analysis. Samples from IHF cats and non-IHF cats were evaluated by Next-generation sequencing to determine their bacterial profiles. The microbial diversity (alpha diversity) was significantly decreased in IHF cats compared to non-IHF cats and the bacterial profile (beta diversity) differed significantly between both groups. Due to the presence of atypical bacterial species in the IHF cats, a link to the IHF as the source of their initial infection was formed. DNA-based bacterial profiling can be a source of probative evidence in future forensic cases.

(Advisor – Becky Morrow) (Evaluator - Lisa Ludvico) (External Reviewer–Nancy Trun)

#### 9:30am BREAK

9:45am Alyssa Hurd

### Optimization of an LC-MS/ MS Method for Separation and Identification of Synthetic Drugs on Prison Mail Samples

Synthetic cannabinoids have surged in popularity over the past decade. This poses a number of problems within the prison system, including the difficulty detecting soaked mail samples as well as the safety risk to both inmates and prison staff. While numerous published methods exist for toxicological purposes, there remains the need for an established method to screen mail samples as they are being sent into the prisons. LC-MS/MS was used to separate and identify synthetic cannabinoids utilizing 7 standards: JWH-018, JWH-073, AM2201, XLR-11, AB-CHMINACA, 5F-ADB, and AM-2233. Ultrasonification was utilized to extract the analytes from the paper substrate, followed by the development of a LC-MS/MS method by varying solvent ratios and optimizing mass spectral parameters. The method was validated by analyzing 47 mail samples recovered from the Allegheny County Jail. This study suggests that a valid and reliable method can be developed given the variety of synthetic cannabinoid classes tested. (*Advisor – Stephanie Wetzel*) (*Evaluator - Mandy Tinkey*) (*External Reviewer – Jeff Beckstead*)

#### 10:15am Kaitlin King

#### The Detection of Curcumin in Turmeric Dietary Supplements via LC/MS-MS

Quality control is a large issue within the dietary supplement market. A selection of five over the counter turmeric dietary supplements will be analyzed for their active ingredient, curcumin, by liquid chromatography tandem mass spectrometry (LC/MS-MS) and be compared to nutrition labels as well as the FDA lethal dose recommendation. The researcher hypothesizes that the concentrations of curcumin will not reflect the amount recorded on the nutrition label but will be within safe dosage. A negative ionization mode was used in tandem with an isocratic flow of acetonitrile and 50% (v/v) acetonitrile in water with 0.1% formic acid for a full run time of five minutes. The peak was found to elute consistently around the three-minute mark. Calibration curves were developed to determine the amount of curcumin present within the supplement. This research will be beneficial to furthering accountability of supplement quality control, as well as providing information for investigations into overdose.

(Advisor – Stephanie Wetzel) (Evaluator - Mandy Tinkey) (External Reviewer – Fred Fochtman)

#### 10:45am Katherine Wodyka

Perception of Dominance of Individuals based on the Variability of Context

Perception is a part of our everyday lives, we look at the people and situations around us to form our own conclusions. Perceptions are a big part of the criminal justice system for example, police officers are often tasked with solving and diffusing situations without the full context. The study's goal is to draw attention to how context can affect people's perceptions. Participants were shown confrontational images between 2 individuals. Some received the scenario with a brief paragraph of context, while others received no context. Participants then rated a character's dominance and explained their choice. Generally, participants with context found the individual starting the confrontation to be more dominant. Participants were then categorized to determine if trends were present. (Advisor – Lyndsie Ferrara) (Evaluator - David Delmonico) (External Reviewer– Megan Overby)

#### 11:15am LUNCH

12:00pm Jared McAtee

The Accuracy of Ammunition with the Addition of Luminescent Markers

The addition of luminescent markers within gunpowder has shown to be a viable technique to enhance the collection of GSR by using a UV Light. The accuracy of ammunition with the addition of luminescent markers was tested on three different firearms at varying weight percent compared to the gunpowder that was used in each firearm. Five samples were loaded in each of the different groups, including a control group where no luminescent markers were added. A T-Test concluded that there was no significant difference between the accuracy of the control groups and any of the samples with the luminescent markers. A second trial was performed by removing gunpowder equal to the amount of luminescent powder that was added. At low amounts of luminescent markers replacing gunpowder there was no significant difference in the accuracy. As more gunpowder was replaced with luminescent markers, the accuracy became significantly worse than the control.

(Advisor – Stephanie Wetzel) (Evaluator - Lyndsie Ferrara) (External Reviewer – Brian Kohlhepp)

#### 12:30pm Lindsay Loughner

Genetic variation in the human relaxin 2 (*RLN2*) promoter affects *in vitro* expression, with potential implications for susceptibility to preterm birth

Relaxin 2 (RLN2) is a hormone produced during pregnancy. Increased RLN2 levels decrease the tensile strength of fetal membranes and can cause early rupture, resulting in preterm birth. Women assaulted during pregnancy are twice as likely to deliver prematurely; thus, it is important to identify pre-existing genetic risk factors in assessing culpability. Previous studies linked serum RLN2 levels and the risk of preterm birth with a single nucleotide polymorphism (SNP rs3758239) in the *RLN2* promoter. We investigated the consequences of variation at SNP rs3758239 and a nearby microsatellite (CT)<sub>n</sub>(GT)<sub>m</sub> within the promoter on expression. Significant differences in *RLN2* transcription between dinucleotide repeat lengths of the microsatellite (p < 0.005) through one-way ANOVA and linear trend analysis were observed. Our data suggest *RLN2* expression increases as the length of the microsatellite increases. Future work will examine the mechanistic basis for differing expression levels among haplotypes, and its clinical and forensic relevance. (*Advisor – Michael Seaman*) (*Evaluator - Lisa Ludvico*) (*External Reviewer– Amanda Zielen*)

1:00pm Evan Penrod

Evaluation of Gut Microbiota from Vermin Bodies Decomposing in a Confined Environment

When determining a Post Mortem Interval (PMI), two main variables are associated with cadaver decomposition: insects and bacteria. Based on quantity and growth phases of necrophagous insects that accumulate in and around the body, entomologists can estimate a PMI. The presence of physical barriers, or anything that can prohibit insects from reaching a body can negatively skew a PMI estimation. The internal bacteria found in the gastrointestinal tract also begin to change at the moment of death and can be utilized in death studies. Human decomposition behind barriers was simulated with feeder mice in lunchboxes. The use of 16s rRNA sequencing was used to analyze the bacteria within the gastrointestinal tract. There were eight orders of bacteria consistent between all samples: Actinomycetales, Bacteroidales, Bacillales, Lactobacillales, Clostridiales, Campylobacterales, Enterobacteriales, and Pseudomonadales. More samples need to be tested to determine any trends related to PMI and prevalence to specific orders of bacteria.

(Advisor – Lyndsie Ferrara) (Evaluator - Lisa Ludvico) (External Reviewer – Christopher Divito)

#### 1:30pm BREAK

1:45pm Erin Monko

Evaluation of Reference Profiles Created Using the Kinship Feature of TrueAllele®

Kinship analysis allows the genetic profile of an individual to be inferred from family member information as a probability distribution. Generally, increasing the amount of familial information available provides more specific profiles and greater possible match statistics. In this study, the TrueAllele<sup>®</sup> probabilistic genotyping system was used to generate profiles for individuals from three families using a varied number of relatives. The specificity of these kinship-inferred profiles was evaluated to determine their distinctiveness from the general population. These profiles were then compared to the true reference profiles of the target and their family members to compute log(LR) match statistics. Based on the amount of familial information used to generate the profile, an average expected match statistic was developed to serve as a frame of reference for the magnitude of those obtained from kinship inferences in casework and allow true matches, other familial matches, or false positives to be differentiated.

(Advisor – Lyndsie Ferrara) (Evaluator - Lisa Ludvico) (External Reviewer – Olivia D. Goodwin)

#### 2:15pm Joshua Kotchey

#### Synthesis of Lanthanide-Doped Gahnite for Use in Fluorescent Detection of Gunshot Residue

Gunshot residue (GSR) is microscopic and not readily visible at a crime scene. A fluorescent additive to gun powder would allow for a quick visualization of GSR during an investigation. An ideal additive would be lanthanide-doped gahnite ( $ZnAl_2O_4$ ). Gahnite is a naturally occurring mineral with high thermal and chemical stability. The crystal structure of gahnite allows for doping of the mineral, by inserting a trace amount of a different element into the host compound. The literature reveals the capability of different lanthanides (III) to be doped into synthetic gahnite with the plus three oxidation state being the most intensely fluorescent. This was achieved by heating stoichiometric mixtures in a programmable furnace, with the target formula of  $ZnAl_{2-x}(III)_xO_4$ . Co-doping lanthanides was also investigated. Future studies will consist of using trace amounts of the synthesized powder for use in ammunition to determine the effects of the additive on trajectory, velocity and accuracy.

(Advisor – Stephanie Wetzel) (Evaluator - Allison Laneve) (External Reviewer – Brian Kohlhepp)

#### 2:45pm Miranda McCune

Analysis of Blood Spatter on Stain Resistant and Non-Stain Resistant Interior Paints

The purpose of this research was to determine if there was a difference in the size and shape of impact bloodstains on interior walls painted with stain resistant paints and non-stain resistant paints. Due to the variety of interior paints available today it is important to determine if the paint needs to be considered when analyzing a crime scene. Five stain resistant paints and five non-stain resistant paints on primed drywall samples were tested using a rat-trap to simulate a reproducible impact spatter. Five replicate blood spatter samples were created for each of the different paint types. Then five representative stains were chosen from varying areas on each sample, and analyzed. The results indicate a negligible difference between the stain resistant and non-stain resistant paints. This research demonstrates that the type of interior paint on walls has minimal effect on blood spatter analysis.

(Advisor – Lyndsie Ferrara) (Evaluator - Benjamin Cooley) (External Reviewer – Brian Kohlhepp)

3:15pm BREAK

#### 3:30pm Kylie Wilson

#### Analysis of Blood Spatter on Different Stain Resistant Sprayed Fabrics

Bloodstain pattern analysis is an important aspect of forensics that is utilized in reconstructing crime scenes. Minimal research has examined bloodstains on porous surfaces, and no research exists analyzing bloodstains on treated fabrics. The purpose of this research was to analyze impact bloodstain patterns on fabrics treated with various types of stain resistant sprays. It was hypothesized that chemical treatment would alter the morphology of bloodstains on fabric. The results showed Scotchgard<sup>®</sup> and Guardsman sprayed fabrics did not absorb bloodstains, whereas Faultless<sup>®</sup> and Kiwi<sup>®</sup> sprayed fabrics acted similar to non-treated fabrics, allowing for significant absorbance. When determining the angle of impact there was no difference in accuracy between the non-sprayed and sprayed fabrics. When the compositions of the fabrics changed, the morphology of the stains also changed. Each spray resulted in different alterations of the fabrics' absorption and porosity as compared to the unsprayed fabrics, which affected the bloodstain patterns.

(Advisor – Lyndsie Ferrara) (Evaluator - Benjamin Cooley) (External Reviewer – Brian Kohlhepp)

4:00pm Shannon Mahoney Extraction and sequencing of human environmental mtDNA from natural, aqueous environments

The identification of human remains belonging to missing persons has been considered a fairly challenging and universal problem in the field of forensic genetics. The application of current DNA protocols to missing persons investigations would provide a more empirical method for the location of persons missing in bodies of running water or victims of certain mass casualty events. In this study, small bodies of running water were utilized to stimulate a natural decay of human epidermal tissue. From the decaying tissue, an expulsion of epidermal cells and other biological matter are collected at different incremental distances downstream, in an attempt to successfully link said downstream material to our source DNA. This study is developed to act as an exploratory study to determine to what extent human environmental DNA (eDNA) can be pulled from natural environments and sequenced utilizing amplification by AFDIL mitochondrial mini-primer sets hybridized with ecological DNA analysis methodologies.

(Advisor – Lisa Ludvico) (Evaluator - Benjamin Cooley) (External Reviewer – Malachi Weaver)

#### 4:30pm Nathan McFadden

Using Cell-Free DNA to Improve STR Analysis of Sweat Samples

Touch DNA (tDNA) samples in the forensic community are known to be difficult samples to obtain ample amount of template DNA and produce DNA profiles using STR analysis. Cell-free DNA (cfDNA) is DNA present outside the cell and found in bodily fluids. Previous research has thought cfDNA could be used to enhance DNA profiles obtained from tDNA samples. However, they have not been conducted with a cell-free DNA extraction kit. Qiagen extraction kits were used to extract DNA from blood serum samples. The two kits used were QIAmp DNA Mini Kit and QIAmp Circulating Nucleic Acid Kit. DNA yields and STR profiles were compared between the two kits and confirmed the Circulating Nucleic Acid kit provided larger amounts of extracted DNA and improved profiles. This process was conducted with sweat samples. If cfDNA can be validated, cfDNA could be a key component in the genotyping of touch DNA samples.

(Advisor – Lisa Ludvico) (Evaluator - Benjamin Cooley) (External Reviewer – David Orbin)

## Graduate Research Symposium Day 2

### Friday, April 12, 2019 Fisher Hall Room 719 10:00am—11:30am

Time	Title	Presenter
10:00AM	Use of IRMPD Spectroscopy to Characterize Derivatives of Aldehydes Considered Emerging Explosive Threat Compounds	Connor Graca
10:30AM	Detection and Identification of Model Peroxide Explosives Using Paper Spray Ionization Combined With Tandem Mass Spectrometry	Madeleine Wood
11:00AM	Development of a Metal Ion Extraction Protocol for GSR in Blood Matrices with Analysis by SEM-EDX	Emma-Rae Ranger

#### 10:00am Connor Graca

Use of IRMPD Spectroscopy to Characterize Derivatives of Aldehydes Considered Emerging Explosive Threat Compounds

Access on the internet to simple chemistry protocols for new classes of explosives has increased demand for their detection in improvised devices. One emerging threat is the use of common cooking spices, which may be used to increase the explosive power of a homemade explosive device. Mass spectrometry is widely used in forensic investigations and explosives detection. Previous work by our group has demonstrated that derivatization of aldehyde constituents of several cooking spice extracts to create imines can increase their ionization efficiency by both electrospray and electron impact (EI) ionization. In this study, derivatized aldehydes from spice extracts were structurally characterized using IRMPD spectroscopy as part of a wider project to map the fragmentation pathways following EI or collision-induced dissociation.

(Advisor – Michael Van Stipdonk) (Evaluator - Stephanie Wetzel) (External Reviewer – Joe Bennett) (Additional Reviewer - Christine Marsh)

10:30am Madeleine Wood

Detection and Identification of Model Peroxide Explosives Using Paper Spray Ionization Combined With Tandem Mass Spectrometry

Paper spray ionization (PSI) combined with tandem mass spectrometry was tested for the characterization of peroxide explosives. A PSI source was constructed and interfaced to a ThermoFisher Scientific LTQ linear ion trap mass spectrometer. Benzoyl peroxide, dicumyl peroxide, methyl ethyl ketone peroxide, and di-*tert*-butyl peroxide were used as surrogates. Methanol, ethanol, acetonitrile and acetone were investigated as solvents, and Na<sup>+</sup>, K<sup>+</sup>, Rb<sup>+</sup> and Ag<sup>+</sup> as cationizing agents. Both swipe sampling and depositing the solution onto filter paper were tested. Dicumyl, benzoyl, and di-*tert*-butyl peroxide yielded abundant intact, metal-cationized species. Formation of metal-adducts to intact peroxides was confirmed by identifying the mass shift when K<sup>+</sup> or Rb<sup>+</sup> was substituted for Na<sup>+</sup>. Semi-quantitative measurements were possible when comparing overall signal intensities from varying concentrations. Swipe sampling showed lower signal intensity than depositing the solution onto the filter paper. This method demonstrates potential for performing rapid analysis for selective screening of peroxide explosives.

(Advisor – Van Stipdonk) (Evaluator - Stephanie Wetzel) (External Reviewer – Theodore Corcovilos) (Additional Reviewer– Joe Bennett)

11:00am Emma-Rae Ranger

Development of a Metal Ion Extraction Protocol for GSR in Blood Matrices with Analysis by SEM-EDX

The purpose of this study is to extract GSR particles from blood matrices to advance the amount of critical evidence that can be collected from crime scenes/suspects. Through the modification of metal ion extractions, GSR particles is extracted and analyzed for direct comparison by the SEM. Without a modified extraction, the SEM-EDX fails to look beyond the iron in the blood for GSR. Samples were obtained by placing blood on the hands of a shooter and sampling after a shot had taken place. This process was repeated with three different pistols: Glock 19, 380 Bodyguard, and an FEG 22. Using the SEM to determine if GSR samples are present, metal ions are extracted from both the plasma and the red blood cells. The red blood cell extraction is performed as not to damage the integrity of the hemoglobin in blood, while the plasma/buffy coating is put through a full metal ion extraction.

(Advisor – Michael Van Stipdonk) (Evaluator - Stephanie Wetzel) (External Reviewer – Mandy Tinkey)