

MFRC

Midwest Forensics Resource Center

**Research and Development
Program Summary**

November 2010

Midwest Forensics Resource Center (MFRC) Research and Development Program

Introduction

The mission of the MFRC Research and Development Program is to provide technological advances in forensic science for the benefit of our regional partners, as well as the forensic community at large. Key areas of forensic science needs are identified through participation in national meetings in forensic science and guidance by national studies and reports. Under the sponsorship of the National Institute of Justice (NIJ), the MFRC solicits proposals for the development of practical and useful forensic science tools that require proof-of-concept experimentation and tools proven in other fields that require experimentation to demonstrate feasibility for addressing specific forensic science needs. The MFRC facilitates proposal development by working to establish partnerships between researchers and our regional partners. The MFRC administers a peer-review of the proposals and then funds the selected projects at a cost of approximately \$55,000 each, with a 12-month period of performance.

The process for selection of these projects includes the following steps: 1) Drafting of a call for proposals by MFRC staff; 2) Review of the draft call by members of the R&D Advisory Committee; 3) Review and approval of the call by NIJ; 4) Issuance of the call to Iowa State University (ISU), Ames Laboratory, regional partners, and various academic and non-academic research organizations; 5) Receipt of proposals; 6) Review of proposals by R&D Advisory Committee; 7) Ranking and selection by MFRC staff using Advisory Committee reviews; 8) Concurrence by NIJ of selected proposals; 9) Notification of proposers; 10) Receipt and review of progress reports by MFRC; 11) Receipt and review of final reports by MFRC, R&D Advisory Committee, and NIJ; and 12) Posting of final reports on the MFRC website.

The decision to fund any specific project is based upon a peer-reviewed call-for-proposal system administered by the MFRC. The reviewers are crime laboratory specialists and scientists who are asked to rate the proposals on four criteria including: 1) Relevance to the mission of the MFRC; 2) Technical approach and procedures; 3) Capabilities, teaming, and leveraging; and 4) Dissemination and implementation of research findings. A successful proposal demonstrates knowledge of the background for the research and includes a research methodology with a well-defined plan to transfer research findings into the hands of stakeholders to pursue further research or into the hands of users to facilitate application of the developed tools.

Program Summary Technical Sheets

The following project summaries, while not a complete summary of all research areas, are meant to demonstrate the range of research funded by the MFRC. The project summaries describe the forensic need the projects serve as well as the benefits derived from the technology. The summaries provide a brief description of the technology and the accomplishments to date. In addition, the collaboration with regional partners and the status of the dissemination of project results and implementation of the product are highlighted. These technical summaries represent the development and implementation of practical and useful technology for crime laboratories that the MFRC hopes to accomplish.

Acknowledgments

The research projects described in this booklet were funded by the U.S. Department of Justice, National Institute of Justice, through the Midwest Forensics Resource Center at Ames Laboratory, under Interagency Agreement number 2008-DN-R-038.

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Abstracts of Funded Projects

CHEMISTRY

Analysis of Automotive Clear Coat Paints by Micro-Laser Raman Spectroscopy

Jay Siegel, School of Science - Indiana University-Purdue-University Indianapolis (IUPUI), Indianapolis, IN

This project partially funds the purchase of a micro-laser Raman spectrometer to evaluate the feasibility and discriminating power of Raman spectrometry in the characterization and analysis of automotive clear coat paints. This represents one phase of an ongoing project in the analysis of automotive clear coat paints. This represents one phase of an ongoing project in the analysis of clear coats undertaken by the Forensic and Investigative Sciences Program at IUPUI. The methods used include micro-laser Raman spectroscopy, Fourier transform infrared spectrophotometry, and UV-visible-near infrared spectrophotometry. The Raman phase of the project is conducted in partnership with the Indiana State Police Forensic Science Lab.

Application of a Novel Mixed-Mode Reversed-Phase HPLC Column to the Rapid Confirmatory Analysis of Intoxicants and their Hydrophilic Metabolites by LC-MS/MS

Dwight Stoll, Department of Chemistry - Gustavus Adolphus College, St. Peter, MN

High Performance Liquid Chromatography (HPLC) has become the dominant analytical methodology in forensic drug analysis. Yet, low retention of highly hydrophilic compounds has historically been a significant weakness of reverse-phased HPLC. This project develops rapid (less than six minutes per analysis) reverse-phased HPLC methods for confirmatory analysis of common benzodiazepines and opiates and their major metabolites in blood and urine. Detection of intoxicants is achieved by liquid chromatography tandem mass spectrometry.

Application of Chemometric Procedures to Differentiate Ignitable Liquid Residues from Substrate Interferences: Creating Reference Collections and Optimizing Data Analysis Procedures

Ruth Waddell-Smith, School of Criminal Justice - Michigan State University, East Lansing, MI

In arson investigations, fire debris is typically extracted and analyzed to determine the presence of ignitable liquid residues. However, identification of these residues can be complicated by interferences from substrates. This project targets development of an objective methodology for differentiating ignitable liquids from burned substrates by creating reference collections. The project also demonstrates the potential of statistical and chemometric procedures to associate and differentiate classes of liquids and to differentiate liquids from burned substrate interferences.

Application of Multivariate Statistical Procedures in Fire Debris Analysis: Investigating Matrix Interference Effects and Weathering of Ignitable Liquids on Association of Ignitable Liquid Residues to Neat Ignitable Liquids

Ruth Waddell-Smith and Victoria L. McGuffin, School of Criminal Justice and Department of Chemistry - Michigan State University, East Lansing, MI

Previous research demonstrated the successful use of principal component analysis and Pearson product moment correlation coefficients for the association and discrimination of ignitable liquids. This project considers the effect of matrix interferences and liquid weathering on ignitable liquid residue association to the Neat liquid.

Comparison of Gas Chromatography-Mass Spectrometry and Liquid Chromatography-Mass Spectrometry for Discrimination of *Salvia divinorum* from Related *Salvia* Species Using Chemometric Procedures

Victoria L. McGuffin and Ruth Waddell-Smith, Department of Chemistry and School of Criminal Justice-Michigan State University, East Lansing, MI

Salvia divinorum is a hallucinogenic herb that is banned in 17 states. The plant is one of nearly 1,000 species of *Salvia*, some of which are culinary herbs and ornamental shrubs. The research develops a method to definitively identify *S. divinorum*. Samples are analyzed by both gas chromatography-mass spectrometry (GC-MS) and liquid chromatography-tandem mass spectrometry (LC-MS/MS) to determine which technique offers the greatest selectivity for the identification of *S. divinorum*. Chemometric procedures are used to identify the most characteristic chemical components that allow differentiation of *S. divinorum* from other *Salvia* species.

Fast Gas Chromatography Capabilities in Drug Identification

Charles Cornett, Department of Chemistry and Engineering Physics, University of Wisconsin-Platteville, Platteville, WI

Fast gas chromatography with hydrogen mobile phase (Fast GC-H2) enables significant increase in sample throughput, chromatographic resolution and budget savings in drug identification involving GC separations. Peer-reviewed research in Fast GC and recent work by the PI with Fast GC-H2 in ignitable liquid identification demonstrated significant improvements in resolution and reductions in retention times. This work will assess Fast GC-H2 in drug identification, provide data for the use of H2 in GC-MS, and create webinar modules and discussion networks for laboratory for laboratory personnel to discuss the potential implementation of Fast GC-H2.

Rapid Arson Sample Analysis Using DART Mass Spectrometry

John McClelland, Ames Laboratory-U.S. Department of Energy, Iowa State University, Ames, IA

Gas chromatography/mass spectrometry (GC/MS) is commonly used to identify accelerants in fire debris, but it is not ideal, requiring substantial time for sample analysis and data interpretation. In collaboration with arson investigators, this project investigates applying a new, faster mass spectrometry (MS) method for arson analysis called direct analysis in real time (DART). DART-MS has the speed of a screening technique and the precision of GC/MS. Project objectives include collecting DART-MS spectra of accelerants and common fire-debris matrices burned with and without accelerants, and demonstrating the identification of accelerants as unknowns from the DART-MS spectra of burned matrices.

DNA

Degradation in Chromosomal DNA Assessed Using PCR Amplification and Capillary Electrophoresis

Robert Allen and Emily VanDegrift, Department of Forensic Sciences, Oklahoma State University, and Byron Smith, Tulsa Police Department Crime Laboratory, Tulsa, OK

DNA profiling methods are widely used in the forensic community. For highly degraded DNA samples, the number of methods is limited, and oftentimes require the use of new, expensive instrumentation. This project investigates the use of quantitative template amplification technology (Q-TAT) assay to provide information about DNA integrity.

DIGITAL EVIDENCE

A Steganalyzer Package for Forensic Applications

Jennifer Davidson, Department of Mathematics/Electrical Engineering - Iowa State University, Ames, IA

The use of steganographic software for hiding information in image files for illicit purposes is becoming more widespread. While steganalysis algorithms are abundant in academic literature, there are few software programs that address the needs of local police departments who perform computer forensic functions for steganalysis. This project extends the proof-of-concept established on the use of an artificial neural network for wavelet steganalysis in an earlier study. Specific aims are to enhance the capabilities of the software and to make it available for practical use in computer forensics.

PATTERN EVIDENCE

A Method for Lifting Bloody Impressions Using a Lifting Strip Containing Titanium Dioxide

Jessica Zarate, Northville City Police Department, Northville, MI

Bloody impressions are of great importance to the forensic community as they are frequently encountered at crime scenes. Impression evidence in blood cannot always be removed from the crime scene for analysis in a laboratory setting, so many potentially identifiable impressions can only be photographed and not enhanced, thereby complicating the latent print identification process. In an earlier study, a lifting strip containing titanium dioxide was successful in lifting and enhancing bloody fingerprints from several non-porous and semi-porous surfaces of contrasting colors. This study improves upon the methods used to lift bloody palm prints, foot prints, and footwear impressions, in addition to bloody fingerprints from porous, semi-porous, and non-porous surfaces.

Application of Face Recognition Technology to Microstamped Cartridge Cases

Scott Chumbley, Department of Materials Science and Engineering - Ames Laboratory, U.S. Department of Energy, Ames, IA. Song Zhang, Department of Mechanical Engineering - Iowa State University, Ames, IA

Microstamping identification has been suggested as a way of providing an objective means to relate guns to fired ammunition. While the method shows promise, questions remain with regard to the durability of the mark. This project determines whether microstamped identifiers can be recognized using a combination of imaging and software recognition methods. Three different quality firearms are obtained and then fired, cleaned, and polished at regular intervals until 1,000 rounds have been fired or the identifier is unreadable. Data is taken using an optical profilometer and analyzed using computers to see if the identifier is recognizable. A corollary study uses metallurgical etching to determine if identifiers purposely removed can be recovered.

Discrimination of Dyed Cotton Fibers Based on UV-Visible Microspectrophotometry and Multivariate Statistical Analysis

John Goodpaster, Department of Chemistry and Chemical Biology - Indiana University Purdue University Indianapolis (IUPUI), Indianapolis, IN

Dyed cotton fibers are a common fiber type found in clothing. One of the most popular methods for their analysis is microspectrophotometry. This project analyzes dyed cotton fibers by microspectrophotometry and evaluated the UV-visible spectra using multivariate statistical analysis. This is done to determine the extent to which spectra can be differentiated, identify discriminating spectral features, assess whether spectra can be shared between laboratories, and examine the potential benefits of coordinate transformations and first derivatives.

Informing the Judgments of Fingerprint Analysts Using Quality Metric and Statistical Assessment Tools

Glenn Langenburg, Minnesota Bureau of Criminal Apprehension Forensic Science Laboratory, St. Paul, MN

Fingerprint analysts typically use a number of tools in their decision-making process. This project evaluates the use of two newly developed tools to determine how information regarding the clarity of friction ridge features (quality measurement tool) and the strength of the corresponding friction ridge features (probabilistic tool) inform the judgements of fingerprint analysts. The two tools are also used to determine how information regarding the strength of the corresponding features (when provided by other fingerprint experts) will inform the judgements of the participating analysts. The measured variables for the effect on examiner performance are the accuracy and reproducibility of the conclusions against the ground truth and analyst variation during feature selection.

Physical Matching in Trace Evidence: A Validation Study Using Automobile Parts

Joseph Wermeling, Wisconsin State Crime Laboratory, Madison, WI. Charles Cornett, Department of Chemistry/Engineering Physics - University of Wisconsin-Platteville, Platteville, WI

Physical matches, or fracture matches, constitute an important procedure in trace evidence analysis. In spite of the widespread use of these processes, few validation studies have been conducted to verify what is considered to be an intuitively obvious process. This study validates the physical matching process of questioned automotive parts to a known vehicle in simulated hit-and-run accidents. Laypersons are utilized to test the hypothesis that a given physical match is indeed an intuitive process and to strengthen the position of the forensic examiner who is viewed as more capable in determining physical matches.

Shape Measurement Tools in Impression Evidence: A Statistical Approach

Mary A. Bush and Peter J. Bush, Laboratory for Forensic Odontology Research (LFOR), School of Dental Medicine, State University of New York (SUNY) at Buffalo, Buffalo, NY H. David Sheets, Department of Physics, Canisius College, Buffalo, NY

This project addresses the need for statistical models in pattern evidence analysis. It builds upon the software developed in an earlier study to examine the principal causes of shape deformation in bite marks. The goal of the project is to explore the feasibility of applying the shape measurement tool to fingerprint evidence and footwear impressions. This will provide forensic practitioners with quantifiable validations of the quality of example items of pattern evidence.

Analysis of Automotive Clear Coat Paints by Micro-Laser Raman Spectroscopy

FORENSIC TECHNOLOGY NEED

Paint evidence is often found at crime scenes involving automobiles. Yet, within the forensic community, little attention has been paid to the analysis of clear coat paints, i.e., the top layer of automotive paints that contains no pigmentation and serves as a protective film. This may be due in part to the lack of inorganics in clear coats, which are almost always present in color or base coats. Recent advances in micro-laser Raman instrumentation make it easier to obtain high quality spectra of small materials, such as the cross-section of a chip of automotive paint.

TECHNOLOGY DESCRIPTION

Micro-laser Raman spectroscopy is a very powerful sampling and analysis technique that acquires spectra of extremely small areas nondestructively and without touching the sample. Although widely used in many different fields, the technique has seen little application by forensic scientists.

METHODOLOGY

The main objective of this research is to evaluate the effectiveness of micro-laser Raman spectroscopy in discriminating among automobile clear coat paints. A secondary objective is to evaluate micro-laser Raman spectroscopy as part of a scheme to analyze automotive clear coats. Other techniques that are used in such a scheme are also evaluated. They include Pyrolysis Gas Chromatography-Mass Spectrometry, Fourier Transform Infrared (FTIR) spectrophotometry, and Ultra-Violet (UV)-Visible-Near Infrared (NIR) Spectrophotometry.

Specific goals to evaluate the feasibility and discriminating power of Raman spectrometry in the characterizing and analyzing of clear coats include:

- Purchase and evaluation of the micro-laser Raman spectrometer
- Optimization of sampling and analysis techniques
- Validation of the techniques and proposed scheme
- Testing the effectiveness of the scheme to analyze automotive clear coats



Figure 1. CRAIG Model CTR-1, 2.5 mW laser at 785 nm with 20x objective.

ACCOMPLISHMENTS AND ONGOING WORK

A CRAIG model CTR-1 micro-laser Raman spectrometer, with a 2.5 mW laser at 785 nm, was purchased for the analysis of clear coats. The instrument was defective and underpowered with spectra plagued by low signal to noise ratio. It was replaced by a similar instrument with a 3.0 mW laser, also at 785 nm. The new instrument is capable of running at 100, 25 or 10% power. Satisfactory spectra were obtained at 10% power.

Two hundred eighty-five samples of automobile paint chips with make and model information were obtained (U.S.-200; Australia-50; U.K.-35), and a method for sectioning the chips was developed. The clear coat was peeled off the paint chip, mounted on aluminum foil, and put on the stage of the Raman microscope.

MAKES (24)		MODELS (106)			
Acura	Subaru	RSX	Tahoe	Ranger	Cherokee Classic
Beretta	Toyota	GTz	Trailblazer	Ranger LT	Wrangler Voyager
BMW		528i	Z28	Taurus	5E Bonneville
Buick		Century	Crossfire	Taurus LX	Rio G6
Cadillac		LeSabre	Lebaron	Taurus SE	Sedona Grand Am
Chevrolet		Rendezvous	Plymouth	Taurus SEL	Sephia Grand Prix SE
Chrysler		Deville	Sebring	Taurus SES	Sportage Sunbird LE
Dodge		Deville DTS	Explorer	Windstar	3 Trans Am
Ford		SRX	4x4	Jimmy	626 xB
GMC		Uplander	Caravan	Sierra	Protégé Silverado
Honda		1500	Dakota	Sonoma	Cougar Camry
Infiniti		Blazer	Durango	Yukon	Marauder Camry DX
Jeep		Cavalier	Intrepid ES	Accord	Sable Corolla
Kia		Cavalier RS	Ram	Accord LX	Eclipse MR-2
Mazda		Cobalt	Ram 1500	Civic	Galant Rav4 4x4
Mercury		HHR	Ram Charger	Civic EX	Mirage Tacoma
Mitsubishi		Impala	Ram Pick-up	Elantra	Altima Breeze
Nissan		Lumina	Escape x LT	Odyssey	Frontier
Oldsmobile		Monte Carlo	F150	Sonata	Sentra
Plymouth		S Series Blazer	F350	G20	Xterra
Pontiac		Santa Fe 2800	F250 x LT	J30	Aurora
Scion		Sebring 4x4	F250 Super DutyV8	Intrigue	
		Outback Sport 4	Touring Town & Country	Grand Voyager SE	

Table 1. Automobile makes and models tested.

To date, 130 samples have been tested under several thickness conditions (whole chip, 15µm, 10 µm, and 5 µm) and spectra were obtained using both 50X and 20X objective lenses. Data revealed that spectra obtained from 50X objective were no better than those from the 20X objective, yet they took longer to collect.

It was also found that 5 µm cross sections generated the clearest Raman spectra. Based on parameters tested, integration has been narrowed to between 20 and 60 seconds, and number of averages between 5 and 10.

Five runs of each clear coat were performed on different parts of the peel. For any given sample, reproducible Raman peaks were obtained, along with consistent results from paint to paint. The vast majority of the samples yielded acceptable

signal to noise ratios. It was decided not to include the ones with a low signal to noise ratio in the chemometric analysis.

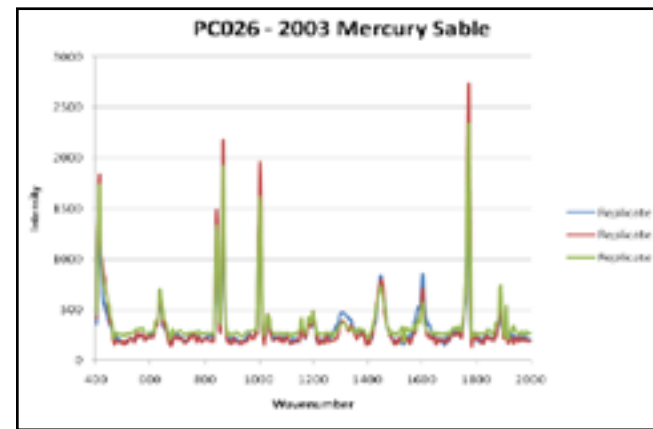


Figure 2. Raman spectrum of a 1992 Mercury Sable clear coat sample showing vibrational energy levels of three replicates.

On two samples, a longitudinal study was performed for eight weeks each. Raman spectra were obtained each week and compared to determine the extent they change over time.

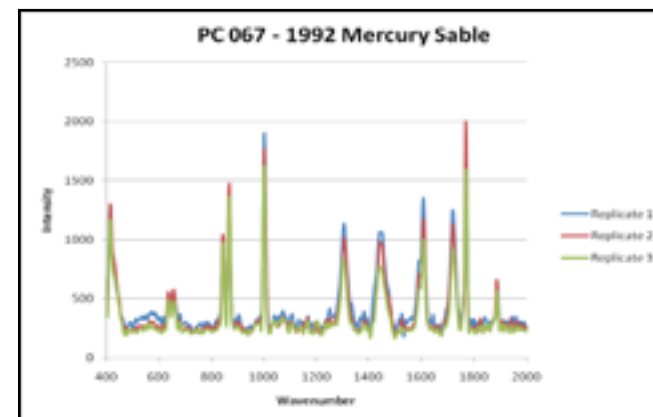


Figure 3. Composite of eight Raman spectra taken weekly of the same sample for a 2002 Pontiac Trans Am.

Currently, the study is still ongoing. When all the samples have been run, Raman spectra will be evaluated by chemometric methods to determine the number of spectra types. Three types of chemometrics will be obtained: Principle Component Analysis (PCA), Agglomerative Hierarchical Clustering (AHC), and Discriminant

Analysis (DA). These analyses are conducted to determine the number of groups the spectra fall into, and to assess how discriminating Raman micro-spectrophotometry is on clear coat analysis.

TECHNOLOGY BENEFITS

Clear coats on automobile paints are very common today. Yet, they have not been analyzed forensically to any degree. This project shows the value of laser Raman spectroscopy when applied to the analysis of trace evidence. Use of the instrument will improve the scientific value of paint evidence and increase the association value of paint evidence to a suspect source.

COLLABORATION

The project is a collaborative effort between Indiana University-Purdue University Indianapolis (IUPUI) Forensic and Investigative Sciences Program and the Indiana State Police (ISP) Forensic Science Laboratory. ISP determines the effectiveness of the scheme and of the micro-laser Raman spectrometer in particular, to characterize and compare known and unknown paint chips. Finally, ISP prepares a number of proficiency tests to use for further validation of the Raman technique and of the scheme.

DISSEMINATION

Research findings were presented at the Midwest Forensics Resource Center's annual meeting and at the American Academy of Forensic Science annual meeting. A manuscript will also be prepared for submittal to the *Journal of Forensic Sciences*. A final technical report on the project and its findings will be posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

- Siegel, J., Goodpaster, J., Cline, B., Foos, R. "Analysis of Automotive Paint Clear Coats by Raman Spectroscopy." Presentation made at the Midwest Forensics Resource Center Annual Meeting, July, 2009, Indianapolis, IN.
- Liszewski, E., Lewis, S., Goodpaster, J., Siegel, J. "Improved Methods for the Discrimination of Automotive Paint Clear Coats Using Microspectrophotometry and Chemometrics." Presentation made at the American Academy of Forensic Sciences 62nd Annual Meeting, February 2010, Seattle, WA.

IMPLEMENTATION

Upon completion of the research, ISP will evaluate use of the scheme developed from this project in the analysis of case samples of paint as they come into the laboratory, and as a supplement to their normal analysis protocols. They will also have use of the Raman instrument and microtome for their case work.

CONTACTS

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Application of a Novel Mixed-Mode Reversed-Phase HPLC Column to the Rapid Confirmatory Analysis of Intoxicants and Their Hydrophilic Metabolites by LC-MS/MS

FORENSIC TECHNOLOGY NEED

Benzodiazepines are some of the most widely used prescribed and abused drugs in Midwestern states. To detect parent drugs and hydrophilic metabolites of certain benzodiazepines, most forensic laboratories currently use Liquid Chromatography-Mass Spectrometry (LC-MS) technology to test for 15 drugs and metabolites in blood and urine samples. However, each run is time consuming (about 30 minutes) and many of the closely related drugs are not well separated. A method is needed in toxicology laboratories to improve the speed of analysis, detection limits, and optimize the separation of mixtures of intoxicants.

TECHNOLOGY DESCRIPTION

Low retention of highly hydrophilic compounds has historically been a significant weakness of reversed-phase HPLC (RPLC), which is the most common mode of liquid chromatography in use today. This weakness has prompted the development of several approaches to increase retention, including the use of conventional cation-exchange materials, ion-pairing reagents as additives in mobile phases used for RPLC, and the more recent development of Hydrophilic Interaction Chromatography (HILIC). This project evaluates the application of a novel mixed-mode RPLC column.

METHODOLOGY

Specific objectives of this project are to:

- Compare the retention of 16 benzodiazepines and nine opiates on two LC-MS HPLC columns; the novel carboxylated Hyper-Crosslinked-C8 (Mixed Mode-C8) column and a conventional reverse-phase (StableBond-C18) column.

- Compare the LC-MS/MS detection limits for the 25 compounds using the novel and conventional reverse phase columns
- Adjust the operational parameters (i.e., column length, particle diameter, flow rate, gradient elution conditions, temperature, etc) to optimize the throughput of the assays
- Evaluate the sensitivity and specificity of the LC-MS/MS assays by analyzing blood and urine extracts and comparing the results with data collected by the MN Bureau of Criminal Apprehension using LC or Gas Chromatography-Mass Spectrometry (GC-MS) for the target compounds
- Evaluate false negative and false positive rates for the new method.

ACCOMPLISHMENTS AND ONGOING WORK

Preliminary work with opiates indicates that separation of the nine target opiates, including two hydrophilic and structurally similar glucuronide conjugates of morphine, is possible through new column chemistry from Supelco which is built upon the increasingly popular shell particle architecture.

The chromatograph below shows the separation of the nine opiates. Separation was completed in less than five minutes, yielding good retention and resolution of the glucuronide conjugates of morphine and excellent resolution of all other peaks except for oxycodone and 6-acetylmorphine. This is well within the range of analysis time specified by the project collaborators at the Minnesota Bureau of Criminal Apprehension (BCA).

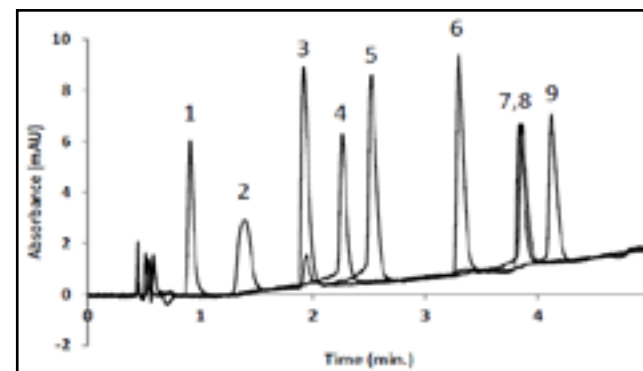


Figure 1. HPLC separation of nine opiate compounds and metabolites using a prototype perfluorinated reversed-phase material built on fused-core particle technology. Compounds: 1-morphine-3- β -D-glucuronide, 2-morphine-6- β -D-glucuronide, 3-morphine, 4-oxymorphone, 5-hydromorphone, 6-codeine, 7/8-oxycodone/6-acetylmorphine, 9-hydrocodone

Although the lack of resolution of oxycodone and 6-acetylmorphine is not ideal, the two compounds are from unrelated parent compounds and thus easily distinguishable by mass spectrometry. The fact that all nine opiates can be separated in a single analysis is an interesting discovery since the two groups of compounds are currently analyzed using two different methods and two different technologies.

Initial work on the benzodiazepine target compound using the same column yielded encouraging results. However, a definitive set of separation conditions was not reached because dealing with 16 compounds in a single analysis was found to be considerably more difficult than dealing with the nine opiates and their metabolites.

Subtle but important changes were noticed in the retention of closely eluting compounds. For example, the pair of compounds in the upper right circle in the insert box of Figure 2 are not separated at all under the condition using salt in the aqueous solvent, whereas there is reasonable resolution of this pair when the salt is added. The reverse is true for the pair of compounds in the lower left circle.

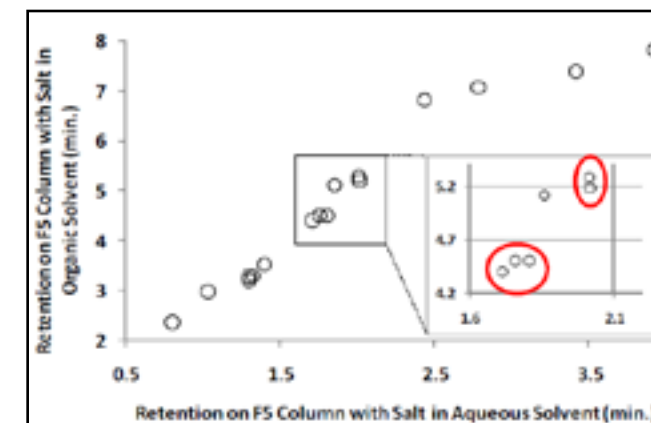


Figure 2. Dependence of the selectivity of 16 benzodiazepines on a perfluorinated reverse-phased column: mobile phase pH buffered by use of 10 mM ammonium formate at pH 2.8 as the aqueous component of the mobile phase (x-axis), and by addition of 100 mM ammonium acetate at pH 4 to the organic component of the mobile phase used in gradient elution (y-axis).

Work is currently continuing on how to control these changes and ultimately choose conditions where overlapping peaks are sufficiently different in terms of parent and fragment masses that further resolution by mass spectrometric detection is straightforward.

TECHNOLOGY BENEFITS

By developing rapid HPLC methods for confirmatory analysis of benzodiazepines, opiates, and their metabolites, this work will greatly improve the screening of drugs of abuse and the throughput in toxicology laboratories.

COLLABORATION

This project is a collaborative effort among Gustavus Adolphus College, the Minnesota BCA Forensic Science Laboratory, and the University of Minnesota. The primary role of the Minnesota BCA is to provide counsel and extracts of blood and urine samples. The University of Minnesota provides the HPLC columns and guidance for their use in this project.

DISSEMINATION

Research findings and results will be presented at annual conferences of the Society of Forensic Toxicologists (SOFT) and the Pittsburgh Conference, and at a meeting of the American Chemical Society. A manuscript will also be submitted to the Journal of Analytical Toxicology and the Journal of Chromatography. Upon completion of the project, a technical report will be posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

- Bonnerup, S., Liskulin, T., Berry, J., Stoll, D. "Development of Rapid LC-MS/MS for Confirmatory Analysis of Opiates and Benzodiazepines". Abstract submitted to the Pittsburgh Conference, March 2011 Pittsburg, PA

IMPLEMENTATION

As opiate analyses potentially can be reduced to a single method, the Minnesota BCA has expressed interest in possible implementation of the technology. To achieve this, the method developed is currently evaluated in accordance with the Minnesota BCA's validation plan for toxicology chromatographic methods. The outcome of the validation process will determine the feasibility of implementation of the Novel Mixed-Mode Reversed-Phase HPLC Column by the Minnesota BCA and other laboratories.

CONTACTS

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Application of Chemometric Procedures to Differentiate Ignitable Liquid Residues from Substrate Interferences: Creating Reference Collections and Optimizing Data Analysis

FORENSIC TECHNOLOGY NEED

In arson investigations, fire debris typically is extracted and analyzed to determine the presence of ignitable liquid residues (ILR). Yet, identification of ILRs can be complicated by interference from substrates, including inherent hydrocarbons and pyrolysis products.

TECHNOLOGY DESCRIPTION

In this project, ignitable liquids, evaporated liquids, and burned household substrates are analyzed to generate reference collections. The potential of chemometric procedures to differentiate ignitable liquids from burned substances is also investigated. The objective is to develop a methodology for the identification of ILRs in the presence of burned substrate interferences.

METHODOLOGY

Specific goals of the research are to:

- Create reference collections of neat and evaporated ignitable liquids
- Create reference collections of burned household substrates
- Assess association and discrimination of neat ignitable liquids, evaporated liquids and burned substrates using Principal Component Analysis (PCA)
- Assess correlations among neat ignitable liquids, evaporated liquids, and burned substrates using Pearson Product Moment Correlation (PPMC) coefficients

ACCOMPLISHMENTS AND ONGOING WORK

Reference collections of 18 liquids from each of the six different classes (as defined by the American Society for Testing of Materials) were created. All liquids were purchased from local stores and internet sources. Examples of common household substrates (nylon, carpet, upholstery, glossy magazine paper, and denim) were also procured from local sources.

Brand Name/Manufacturer	Type of Ignitable Liquid	ASTM Class
Meijer	Gasoline	Gasoline
Shell	Gasoline	Gasoline
Mobil	E85 Gasoline	Gasoline
Goo Gone	Candle wax lifter	Alkane
Lamplight	Ultra-pure paraffin lamp oil	Alkane
Northern Lights	Ultra-pure lamp fuel	Alkane
Goof Off	Ultimate remover	Aromatic
Recochem	Lacquer thinner	Aromatic
Ortho	Insect spray	Aromatic
Sunnyside	Odorless paint thinner	Isoparaffinic
Crown	Odorless paint thinner	Isoparaffinic
Scotch Guard	Fabric and upholstery protector	Isoparaffinic
Tiki	Torch fuel	Naphthalenic paraffinic
Medallion	Lamp oil	Naphthalenic paraffinic
Pennzoil	Marine fuel cleaner and stabilizer	Naphthalenic paraffinic
STP	Fuel injector and carburetor	Petroleum distillate
Meijer	Kerosene	Petroleum distillate
Sunoco	Diesel	Petroleum distillate

Table 1. List of 18 ignitable liquids included in reference collections.

Each liquid was evaporated to four different levels (5%, 10%, 20%, and 50%) by volume. All liquids were spiked onto kimwipes that were extracted for four hours at 80 degrees C using a passive headspace extraction with activated carbon strips. Samples of unburned and burned substrates were also extracted using the same procedure. The carbon strips were eluted with dichloromethane and the extracts were analyzed by GC-MS.

All neat and evaporated liquids were considered as one data set and retention time aligned. Two different retention time algorithms were investigated: a peak matching algorithm and a commercially available correlation optimized warping (COW) algorithm. Optimal alignment was achieved using the COW algorithm (warp 4 and segment size 60) followed by the peak matching algorithms (window size 2).

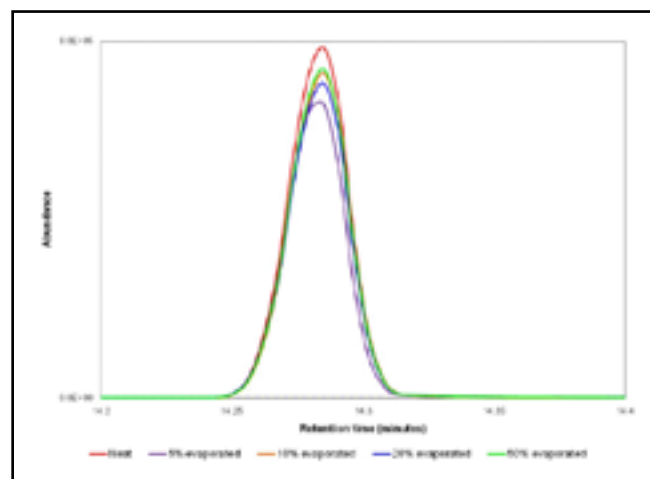


Figure 1. Magnified view of lamp fuel chromatograms (one replicate from neat and 4 levels of evaporation), highlighting the C14 peak aligned using (A) COW algorithm with warp 4 and segment size 60 and (B) COW algorithm with warp 4 and segment size 60 followed by peak matching algorithms with window size 2.

To illustrate proof-of-concept, PCA and PPMC data analysis procedures were conducted on a subset of the ignitable liquids representing each of the ASTM classes in the reference collection. It was found that evaporated liquids can be associated to the corresponding neat liquid, even at evaporation levels of 50%.

To determine if discrimination of matrices and liquids is possible, PCA was conducted and PPMC coefficients were calculated for samples that were not readily distinguished using PCA alone. Based on PPMC coefficients, there was no correlation between burned substrates and liquids. It was concluded that with the combination of PCA and PPMC coefficients, evaporated liquids can be associated to the corresponding neat liquid with differentiation from burned matrices.

Pair-wise comparison	Mean PPMC coefficient ± Standard deviation (n=9)	Strength of correlation
Burned denim vs. Neat lacquer thinner	-0.0045±0.0002	None
Burned denim vs. 50% evaporated lacquer thinner	-0.0044±0.0002	None
Burned upholstery vs. Neat lacquer thinner	-0.0154±0.0003	None
Burned upholstery vs. 50% evaporated lacquer thinner	-0.0151±0.0002	None
Burned magazine vs. Neat gasoline	0.0283±0.0386	None
Burned magazine vs. 10% evaporated lamp fuel	-0.0041±0.0039	None
Burned magazine vs. 20% evaporated kerosene	0.0909±0.0391	None

Table 2. Average PPMC coefficients and standard deviation for liquids and matrices positioned closely in the scores plot for the six neat liquids.

TECHNOLOGY BENEFITS

The creation of a reference collection of burned household items will improve forensic arson investigations. Also, with the developed mathematical procedures, subjectivity associated with the comparison of chromatograms will be improved and objective distinction of ignitable liquids from substrate interferences increased. This will reduce the risk of false positive identifications of liquids in fire debris.

COLLABORATION

This is a collaborative effort between the Michigan State University (MSU) and the Michigan State Police (MSP). Troy Ernst, of the MSP-Grand Rapids Trace Evidence Unit, serves as consultant on the development of the methodologies and their implications to forensic crime laboratories.

DISSEMINATION

The findings of the research have been discussed with potential users and also have been presented at national meetings. A manuscript on the project and its findings has been submitted to the Journal of Forensic Sciences. A final technical report on the research, along with a DVD of reference collections, has been posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

- Baerncopt, J., McGuffin, V., Waddell-Smith, R. "Discrimination of Ignitable Liquids from Burned Matrix Interferences Using Chemometric Procedures." Presentation made at the American Academy of Forensic Sciences 61st Annual Meeting, February 2009, Denver, CO.
- McIlroy, J. "Statistical and Chemometric Analysis of Ignitable Liquids for Arson Investigations." Presentation made to the Illinois State Police, March 2009, Chicago, IL.
- Van De Mark, T., Bodnar, M., McGuffin, V., Waddell-Smith, R. "Association of Evaporated Ignitable Liquids to Their Neat Counterparts Using Pearson Product Moment Correlation Coefficients and Principal Analysis." Poster presentation at the Joint Meeting of the Midwestern, MidAtlantic, Southern, and Southeastern Associations of Forensic Scientists, October, 2009, Orlando, FL.
- Van de Mark, T., Bodnar, M., McGuffin, V., Waddell-Smith, R. "Effects of Matrix Interferences on the Identification of Mixed Ignitable Liquids in Fire Debris Using Chemometric Procedures." Presentation made at the American Academy of Forensic Sciences 62nd Annual Meeting, February 2010, Seattle, WA.

- Baerncopt, J., McGuffin, V., Waddell-Smith, R. "Association of Ignitable Liquid Residues to Neat Ignitable Liquids in the Presence of Matrix Interferences Using Chemometric Procedures" Manuscript accepted for publication in the Journal of Forensic Sciences.

IMPLEMENTATION

The research established proof-of-concept. The investigators are currently applying statistical procedures to assess the effect of matrix interferences and weathering on the association of ignitable liquid residues to neat ignitable liquids (see project summary page 14).

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Application of Multivariate Statistical Procedures in Fire Debris Analysis: Investigating Matrix Interference Effects and Weathering of Ignitable Liquids on Association of Ignitable Liquid Residues to Neat Ignitable Liquids

FORENSIC TECHNOLOGY NEED

In arson investigations, fire debris is typically extracted and analyzed to determine the presence of Ignitable Liquid Residues (ILRs). However, the identification of ILRs can be complicated by interference from substrates and weathering thereby making the association of ILRs to Neat Ignitable Liquids (NILs) very difficult.

TECHNOLOGY DESCRIPTION

Previous research demonstrated the success of PCA (Principal Component Analysis) and PPMC (Pearson product moment correlation) coefficients for the association of ignitable liquids (including evaporated liquids) from the same ASTM class with differentiation of ignitable liquids from different ASTM classes.

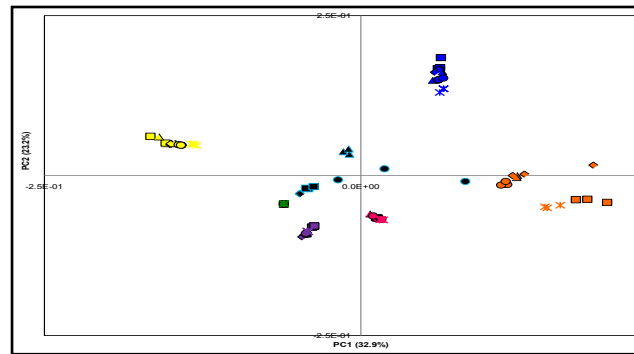


Figure 1. Scores plot of PC1 v PC2 based on the TIC (Total Ion Chromatograms) for six ignitable liquids with scores projected for evaporated liquids and burned substrates. Each color represents a different ASTM class.

This project further develops the objective methodology by considering the effect of both matrix interferences and liquid weathering on ILR association to the neat liquid. Additionally, a Soft Independent Modeling of Class Analogy (SIMCA)

approach for classification of an ILR to a liquid class is investigated, aiming to allow classification with statistical confidence.

METHODOLOGY

The primary objective of this project is to develop a methodology to associate ILRs to the corresponding NILs in the presence of matrix interferences and weathering effects.

Specific goals for the project are to:

- Investigate increased matrix interference effects from four household matrices currently in the “in house” reference collection
- Investigate the effect of increased matrix interference effects on the association of evaporated liquids to neat liquids using PCA and PPMC coefficients
- Investigate the association of simulated ILRs to the corresponding neat liquid using PCA and PPMC coefficients
- Investigate a SIMCA approach for the classification of a simulated ILR to an ignitable liquid class

ACCOMPLISHMENTS AND ONGOING WORK

This is a new project that has not started work yet.

TECHNOLOGY BENEFITS

By developing a methodology for associating ILRs to the corresponding NILs, this project

addresses the National Academy of Sciences concern (*Strengthening Forensic Sciences in the United States: A Path Forward*, 2009) on the ability of many forensic techniques to demonstrate a connection between evidence and a specific individual source. If successful, fire debris analysts, despite the presence of matrix interferences and weathering effects, will be able to attribute statistical confidence in their ILR identifications.

COLLABORATION

The project is a collaborative effort between Michigan State University (MSU) and the Michigan State Police (MSP). The MSP Grand Rapids Trace Evidence Unit serves as a consultant to the development of methodologies applicable to forensic science and useful to the forensic community in the analysis of fire debris.

DISSEMINATION

The results of this project will be presented at the annual meetings of the Midwestern Association of Forensic Scientists and the American Academy of Forensic Sciences. If warranted, a manuscript will be submitted for publication in the *Journal of Forensic Sciences*. Upon completion of the project, a technical report on the project and its findings will be posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

This is a new research project with no publications or presentations to date.

IMPLEMENTATION

Method development is essential to making the research more applicable in the routine casework presented to fire debris analysts. Upon completion of the work, an on-line seminar (accessible through MSU’s on-line learning system) will be developed to educate fire debris analysts on the use of statistical procedures for arson investigations. Developing

and presenting a workshop on training fire debris analysts more thoroughly in the data analysis and interpretation procedures will also be pursued.

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Comparison of Gas Chromatography-Mass Spectrometry and Liquid Chromatography-Mass Spectrometry for Discrimination of *Salvia divinorum* from Related *Salvia* species using Chemometric Procedures

FORENSIC TECHNOLOGY NEED

Salvia divinorum is a hallucinogenic herb which is currently banned in 17 states. With legislation pending in several others, there is a need to identify *S. divinorum* from other *Salvia* species, some of which are culinary herbs and ornamental shrubs. This project develops a method to differentiate *S. divinorum* from the nearly 1,000 species of *Salvia* that are currently known using both chromatographic techniques and chemometric procedures.

State	Classification	Substance Regulated
Florida, Illinois, Kansas, Missouri, Nebraska, North Dakota, Ohio, Oklahoma, and Virginia	Schedule I	Salvinorin A and <i>S. divinorum</i>
Delaware and Mississippi	Schedule I	<i>S. divinorum</i>
South Dakota	Class 1 misdemeanor/ Class 6 felony	Salvinorin A and <i>S. divinorum</i>
Tennessee	Class A misdemeanor	Salvinorin A
Louisiana and North Carolina	Class 3 misdemeanor	Salvinorin A and <i>S. divinorum</i> (only if intended for human consumption)
California and Maine	Class E misdemeanor	Salvinorin A and <i>S. divinorum</i> (only if sold to minors)

Table 1. *Salvia divinorum* regulatory status table.

TECHNOLOGY DESCRIPTION

Previous research investigated the selectivity of different organic solvents for extraction of salvinorin A (the active hallucinogenic component) from dried leaves of *S. divinorum* and *S. officinalis*. Methyl chloride appeared the most promising, extracting the greatest number of components and the highest amount of salvinorin A in *S. divinorum*.

METHODOLOGY

The objective of this project is to build upon the findings in previous work and to develop analytical

methods for the definitive identification of *S. divinorum*, differentiation of *S. divinorum* from other *Salvia* species, and association of adulterated plant matrices with *S. divinorum*.



Figure 1. *Salvia divinorum* plant (left) and dried leaves (right)

This is accomplished by:

- Investigating and comparing three different extraction procedures for components of interest (manual agitation, rotary agitation, and sonication)
- Investigating the selectivity offered by Gas Chromatography-Mass Spectrometry (GC-MS) in creating a chemical fingerprint of *S. divinorum*
- Investigating the selectivity offered by Liquid Chromatography-Mass Spectrometry (LC-MS) in creating a chemical fingerprint of *S. divinorum*
- Comparing chemical markers and using chemometric procedures to identify the most characteristic chemical components that allow differentiation of *S. divinorum* from other *Salvia* species

ACCOMPLISHMENTS AND ONGOING WORK

Findings of previous research were used to optimize

the extraction time for manual and rotary agitation extraction procedures. For manual agitation, the optimal extraction procedure consisted of a 5 minute extraction of the dried plant leaves in dichloromethane. The developed procedure was then used for the extraction of *S. divinorum* and four other *Salvia* species.

Differentiation of *S. divinorum* from other *Salvia* species was investigated by visual comparisons of dried leaves and GC-MS total ion current (TIC) chromatograms. Although this method yielded some results, it was found that quantifiable measures of accuracy and reliability of evidence are needed the most. As a result, chemometric methods were used to differentiate between the five *Salvia* species studied.

Before analyses were conducted, the data collected was pretreated to remove non-chemical sources. Pre-treatment consisted of background subtraction, smoothing, retention time alignment, and normalization. Successful differentiation of *S. divinorum* from other *Salvia* species was achieved using Principal Component Analysis (PCA) with Euclidian distances.

	Average Score on PC1	Average Score on PC2	Euclidean Distance to <i>S. divinorum</i>
<i>S. divinorum</i>	-4.00 ± 2.23 × 10 ⁴	-8.27 ± 0.15 × 10 ⁵	
<i>S. officinalis</i>	-1.32 ± 0.19 × 10 ⁵	1.63 ± 1.28 × 10 ⁵	1.62 × 10 ⁶
<i>S. guaranitica</i>	5.94 ± 1.30 × 10 ⁵	-3.53 ± 0.62 × 10 ⁵	7.92 × 10 ⁶
<i>S. nemerosa</i>	3.24 ± 0.43 × 10 ⁵	4.47 ± 0.45 × 10 ⁵	1.33 × 10 ⁶
<i>S. splendens</i>	4.42 ± 1.13 × 10 ⁵	5.70 ± 0.94 × 10 ⁵	1.48 × 10 ⁶

± one standard deviation n=9

Table 2. Euclidian Distance of *Salvia* Species.

Chemometric methods were also used to determine the association of *S. divinorum* to adulterated plant matrices. The adulteration method consisted of extracting 15.6 grams of *S. divinorum* in acetone for 5 minutes, spiking it onto 0.8 grams of each of the matrices tested in a 10:1 ratio, evaporating the sample to dryness, extracting in triplicate using optimized methods, and analyzing the sample by GC-MS. The matrices tested included *S. divinorum*, *S. officinallis*, *S. sativa*, and *N. tabacum*.

Prior to conducting the analyses, the data were consistent of background subtraction, smoothing, reaction time alignment, and normalization. Successful association of *S. divinorum* to adulterated plant matrices was achieved by PCA with Euclidian distances.

Although the project has yielded some interesting results to date, the project is still ongoing. Currently, the researchers are targeting the development and optimization of analytical methods, the differentiation of *S. divinorum* from other *Salvia* species, the association of adulterated plant matrices to *S. divinorum* by LC-MS/MS and MSⁿ (multiple-stage mass spectrometry), and the comparison of GC-MS, LC-MS/MS and MSⁿ for differentiation and association of *S. divinorum*.

TECHNOLOGY BENEFITS

Findings of the research can assist in the development of optimized procedures for the analysis and identification of *Salvia divinorum*. In doing so, the procedures developed and validated eventually can be implemented in states that have already regulated the plant. They can also be made available to other forensic laboratories should *S. divinorum* or its active ingredient (salvinorin A), become federally controlled in the United States.

COLLABORATION

The project is a collaborative effort between Michigan State University and the Michigan State Police (MSP). MSP provides counsel for analysis.

DISSEMINATION

Project findings and results were disseminated at annual meetings of the American Academy of Forensic Sciences and the Midwestern Association of Forensic Scientists. A manuscript was also submitted to the *Journal of Forensic Sciences*. A final technical report was posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

- Bodnar, M., McGuffin, V., Waddell-Smith, R. "Forensic Analysis of *Salvia divinorum* and Related Salvia Species Using Chemometric Procedures." Presentation made at the 62nd Annual Meeting of the American Academy of Forensic Sciences, February 2010. Seattle, WA.
- McGuffin, V., Waddell-Smith, R., Bodnar, M., and Bugeja, M. "GC-MS and LC-MS/MS for Discrimination of *Salvia divinorum* and Related Salvia Species." Presentation made at the Midwest Forensics Resource Center Annual Meeting, May 2010. Mackinaw Island, MI.
- Bugeja, M., Bodnar-Willard, M., McGuffin, V., Waddell-Smith, R. "Comparison of Methods for the Extraction of Volatile compounds from *Salvia divinorum*." Poster presentation at the Midwestern Association of Forensic Scientists Annual Fall Meeting, October 2010, Kansas City, KS.
- Bugeja, M., Bodnar-Willard, M., McGuffin, V., Waddell-Smith, R. "Comparison of Methods for the Extraction of Volatile Compounds from *Salvia divinorum* Using Liquid Chromatography-Mass Spectrometry for Association and Discrimination from Related Salvia Species." Abstract submitted for presentation at the 63rd Annual Meeting of the American Academy of Forensic Sciences, February 2011. Chicago, IL.
- Bodnar-Willard, M., McGuffin, V., and Waddell-Smith, R. "Forensic Analysis of *Salvia divinorum* and Related Salvia Species Using Chemometric Procedures." Manuscript submitted for publication in the *Journal of Forensic Sciences*.

IMPLEMENTATION

The researchers are currently planning a symposium to present the method developed for extracting salvinorin A in *Salvia divinorum* and the data analysis methods to drug analysts of the Michigan State Police Forensic Science Division.

A copy of the extraction procedures for *Salvia divinorum* was also distributed to the Office of the Attorney General, Crime Laboratory Division, in Bismark, North Dakota.

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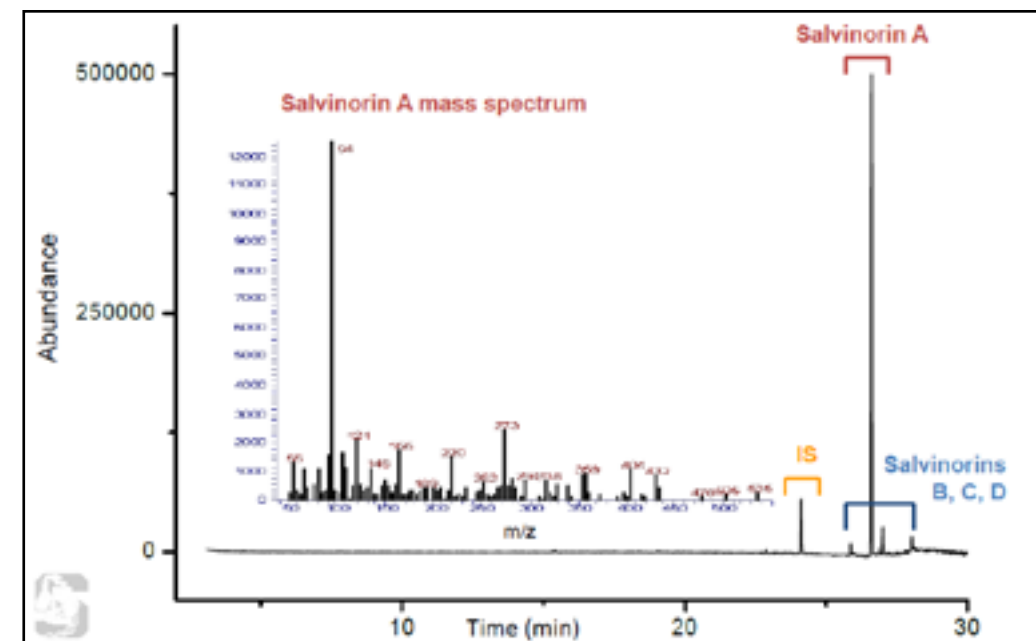


Figure 2. Retention Time Alignment.

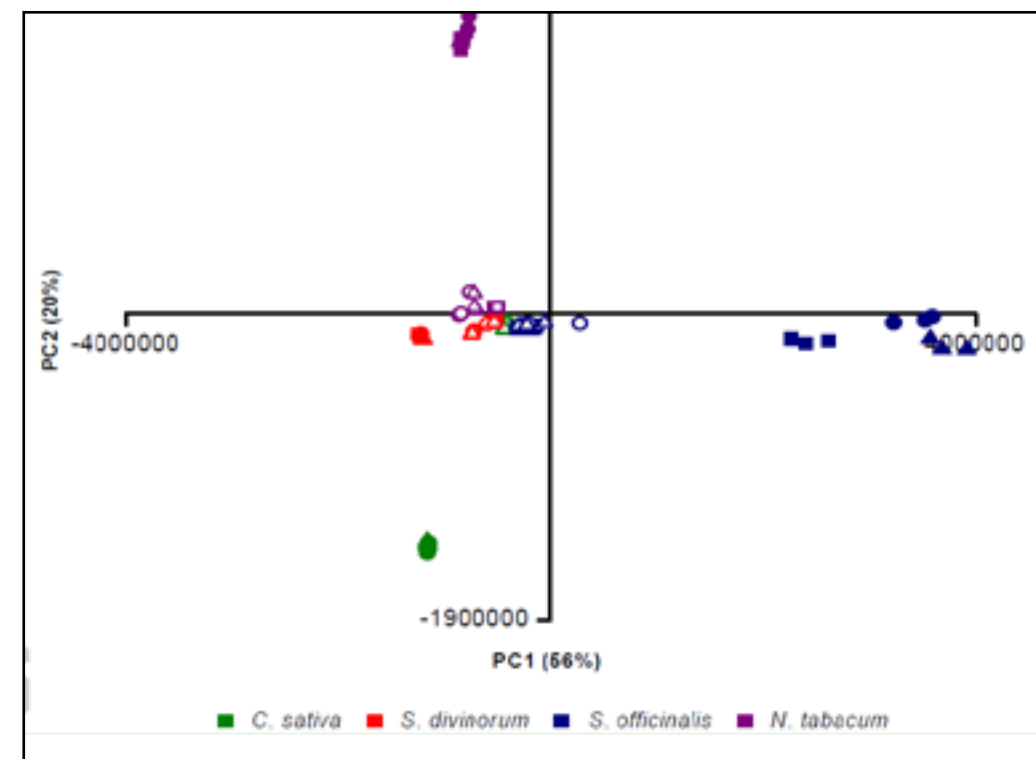


Figure 3. Scores Plot of Adulterated Plant Matrices.

Fast Gas Chromatography Capabilities in Drug Identification

FORENSIC TECHNOLOGY NEED

Gas chromatography (GC), coupled with Mass Spectrometry (MS), is the primary method used at crime laboratories for the identification of drugs. While the technique has sufficient sensitivity and specificity, it can be slow thus limiting the number of samples that can be analyzed. Recent studies indicate that Fast Gas Chromatography (Fast GC) provides a more rapid analysis than conventional GC, especially when combined with hydrogen as a carrier gas. This project examines the capabilities of Fast GC-H2 in drug identification.

TECHNOLOGY DESCRIPTION

Fast GC is a separation technique that couples the stable, rapid heating cycles of Fast GC with narrower capillary columns and high phase ratio. The combination creates more theoretical plates per meter, and enables fast separations with potentially superior resolution and budget savings if hydrogen is used in the mobile phase.

METHODOLOGY

The research builds upon the findings of a similar project funded by the MFRC to examine the capabilities of Fast GC in arson debris analysis. That project demonstrated reductions of more than 50% in retention times of ignitable liquids from arson debris. It also found that the more compressible hydrogen carrier produced improvements in resolution.

While sample preparation time intervals limited the benefits of the Fast GC-H2 combination in ignitable liquids analyses, no such limitations exist in drug analysis procedures and protocols. The goal of this project is to assess the merit of the Fast GC-H2 combination in the identification of drugs.

Specifically, this project assesses:

- The expected gain in resolution and sample throughput for a drug identification unit using a combination of Fast GC and hydrogen carrier gases
- The effect of hydrogen use in GC on mass spectrometry data
- The feasibility of implementing and using the Fast GC-H2 combination at the crime laboratory in light of hydrogen safety with both Mass Spectrometry and Flame Ionization Detection applications.

ACCOMPLISHMENTS AND ONGOING WORK

This is a new project that has not yet started.

TECHNOLOGY BENEFITS

The Fast GC-H2 approach is a relatively easy, simple to implement, and presents great benefits in terms of significantly increasing the instrumental capacity of drug chemistry units, and improving the detection of suspected drugs through better resolution. In addition, the use of hydrogen as a carrier gas may result in significant cost savings over the use of a helium mobile phase.

COLLABORATION

The project is a collaborative effort between the University of Wisconsin-Platteville (UWP) and the Wisconsin State Crime Laboratory at Milwaukee (WSCL-Milwaukee). The WSCL-Milwaukee serves as co-principle investigator in the research project and provides student intern oversight along with controlled substance analysis and research results review.

DISSEMINATION

Research results and findings will be presented at annual meetings of the Midwestern Association of Forensic Scientists and the American Academy of Forensic Sciences. Additional dissemination includes manuscript submission of meritorious results to a peer-reviewed journal, like the *Journal of Forensic Sciences*. Upon completion of the project, a technical report on the project and its findings will be posted on the MFRC website.

Researchers will also investigate dissemination of research findings and results through the online Microgram posting of the U.S. Department of Justice Drug Enforcement Administration. Microgram assists and serves forensic scientists concerned with the detection and analysis of suspected controlled and other abused substances for forensic/law enforcement purposes.

PUBLICATIONS AND PRESENTATIONS

This is a new project with no presentations or publications to date.

IMPLEMENTATION

Switching from a conventional GC to a Fast GC is a relatively easy process. It involves upgrading the power source to either 220 V or 240 V (preferred) service, and installing a high-heat oven shroud, power cord, and narrower capillary column. The cost of switching is about \$4,000. If the results of the research warrant further investigation, the WSCL-Milwaukee wants to conduct an in-house examination of Fast GC-H2 prior to making a switch.

Through a series of asynchronous “webinars”, the UWP will provide training on the use of Fast GC-H2. Videos will be made available through the UWP Media Technology Services Department for analysts to watch at their convenience. Questions on the use of the technology and consultation on

implementation of the approach will also be made available through UWP, along with the opportunity for crime laboratory analysts to travel to UWP to have their samples analyzed using the Fast GC-H2 method.

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Rapid Arson Sample Analysis Using DART Mass Spectrometry

FORENSIC TECHNOLOGY NEED

Gas Chromatography-Mass Spectrometry (GC/MS) is the primary analytical method used in arson investigations. Yet, the technology is not ideal, requiring substantial time for sample analysis and data interpretation. This limits the number of samples that can be tested and subsequently increases the backlog of cases. Subsequently, there is a need for a technology which maintains sensitivity and specificity yet significantly reduces sample time analysis and data analysis complexity.

TECHNOLOGY DESCRIPTION

Direct analysis in real time (DART) is a new ionization method for the rapid detection of analytes. By using mass spectrometry, the ionization source enables near instantaneous determination of sample composition without sample preparation and extraction steps. The technology is versatile and potentially can be applied to a wide range of chemicals.

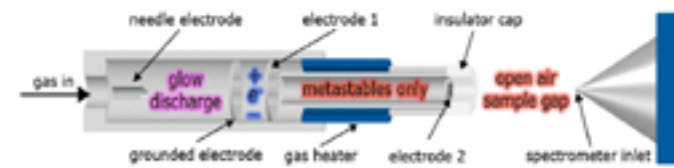


Figure 1. Schematic of the DART ion source (JEOL USA, Inc.).

METHODOLOGY

This project investigates the technology's application to arson sample analysis. Specifically investigators will:

- Collect DART-MS spectra of vapors from the major classes of ignitable liquids to build a small, proof-of-principle library. Representative accelerants from all ASTM classes will be examined.

- Collect DART-MS spectra of common fire-debris matrices after being burned with and without accelerants. Several representative substrate materials (e.g., carpet, wood, and drywall) will be used as arson-sample matrices. Spectra will be added to the library.
- Use the library of data generated to demonstrate the ability of DART-MS to identify accelerants on burned materials. This demonstration is achieved by searching data from the analysis matrices burned with accelerants against the library and matching it to spectra of the accelerants used in burning a sample.

ACCOMPLISHMENTS AND ONGOING WORK

This is a new project that has not started work yet.

TECHNOLOGY BENEFITS

Technology benefits to the forensic science community are three fold: 1) DART-MS can be marketed as an efficient alternative to GC-MS; 2) the ability of DART-MS to analyze samples with great sensitivity, yet without sample preparations, may increase sample throughput at the crime laboratories without sacrificing quality; 3) reduced turnaround times of DART-MS should lead to improved analyses and indirectly to more thorough sampling at fire scenes.

COLLABORATION

This project is a collaborative effort between the Ames Laboratory of the U.S. Department of Energy, the Minnesota Bureau of Criminal Apprehension (BCA), and the Iowa Department of Public Safety (DPS). The Minnesota BCA and the State Fire Department of the Iowa DPS will provide advice on the proper methods for production and handling of test samples and on the data required for successful

investigations. The State Fire Department of the Iowa DPS may also provide samples for analysis from test burns at the Iowa Fire Service Training Bureau.

DISSEMINATION

Research findings and results will be disseminated through presentations at the fall meeting of the Midwestern Association of Forensic Scientists, and through publication in the Journal of Forensic Sciences. Upon completion of the project, a technical report on the project and its findings will be posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

This is a new research project with no publications or presentations to date.

IMPLEMENTATION

The project establishes proof-of-concept for Rapid Arson Sample Analysis Using DART Mass Spectrometry. If successful, the investigators will apply for federal funding to continue the work. Yet, at the completion of the current project, the investigators plan to host a workshop on DART-MS applications in forensic arson analysis.

Full implementation of project findings will probably occur during or at the end of the follow-up project. This may take the form of developing standard operating procedures, submitting data to the forensics database being developed by RTI International (see <https://www.forensicdb.org/>), and through feasibility studies of the DART-MS technology at various crime scene laboratories.

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Degradation in Chromosomal DNA Assessed Using PCR Amplification and Capillary Electrophoresis

FORENSIC TECHNOLOGY NEED

DNA profiling methods are widely used in the forensic community. For highly degraded samples the number of methods is limited and oftentimes differ in discriminating power and tolerance level.

Additionally, some of the methods also require the use of new, expensive instrumentation.

A cost effective method is needed for the efficient identification of usable samples for DNA profiling. This project investigates the use of Q-TAT (quantitative template amplification technology) assay to provide information about DNA integrity.

TECHNOLOGY DESCRIPTION

Previous work resulted in a multiplex PCR (polymerase chain reaction) assay (Q-TAT) that provides quantitative and qualitative information about DNA samples. This project builds upon that work and investigates the ability of the assay to provide information about DNA integrity.

METHODOLOGY

The overall objective of the proposal is to establish parameters suitable for using the Q-TAT assay to reliably identify DNA samples. The samples are sufficiently degraded to require specialized testing methods to produce a DNA profile.

Specific goals of the project are to:

- Identify specific conditions needed to degrade both nDNA (nuclear DNA) and mtDNA (mitochondria DNA) in a controlled fashion using chemical and enzymatic means and to confirm the extent of degradation in a forensically useful way.

- Correlate ratios of fluorescence in large and small amplicons produced in the Q-TAT assay with fluorescence incorporated into STR (short tandem repeat) amplicons for high and low molecular weight loci and for the HVI (hypervariable regions) and HVII amplicons from mtDNA.
- Demonstrate the relationship for fluorescence in Q-TAT amplicons with that seen in STR profiles produced from previously tested evidence samples from the project collaborator.

ACCOMPLISHMENTS AND ONGOING WORK

This is a new project that has not yet started work.

TECHNOLOGY BENEFITS

The benefit of the Q-TAT assay to the forensic community is two fold. First, the method provides a rather complete picture on the quantity and quality of DNA recovered from evidence. Second, a cost effective method for efficient identification of samples for DNA profiling enhances the efficiency of casework processing.

COLLABORATION

The project is a collaborative effort between the DNA Laboratory of the Oklahoma State University's (OSU) Department of Forensic Sciences and the Tulsa Police Department (TPD). TPD provides adjudicated samples for re-testing with Q-TAT as well as DNA data from the samples for re-analysis of Q-TAT amplicon rfu ratios. Additionally, TPD provides validation and casework experience as the TPD laboratory routinely utilizes the assay for DNA quantification.

DISSEMINATION

Project results and findings will be presented at regional forensic meetings and national conferences including the Midwestern Association of Forensic Scientists (MAFS) or the Southwestern Association of Forensic Scientists (SWAFS) Annual Meeting, and the American Academy of Forensic Sciences (AAFS) Annual Conference.

A manuscript on the project and its findings will also be submitted for publication in a peer-reviewed journal. Upon completion of the project, a technical report on the project and its results will be posted on the MFRC website. The possibility of posting research findings and results using OSU distance learning capabilities will also be explored.

PUBLICATIONS AND PRESENTATIONS

This is a new project that has not yet started work.

IMPLEMENTATION

Upon completion of the project, the partners will host two training sessions at their shared Forensic Sciences and Biomedical Research Facility on the OSU campus in Tulsa, OK, to give potential users a chance for hands-on experience using the Q-TAT method. Free training sessions will be offered.

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A Steganalyzer Package for Forensic Applications

FORENSIC TECHNOLOGY NEED

The use of steganographic software for hiding information in image files is becoming more widespread. While steganalysis algorithms are abundant in the academic literature, few address the needs of local police departments performing computer forensic steganalysis. Those currently available are very expensive and require extensive training of forensic personnel.



Figure 1. Message hidden in image (“Attack at Midnight”).

TECHNOLOGY DESCRIPTION

Steganalysis is a procedure that determines with a success rate better than random guessing, whether or not an image contains a hidden payload. In this project, the use of blind steganalysis (general image measures) is pursued to statistically determine a hidden payload in an image. It is based on the principal of the more bits that change value from the original (innocent) image, the easier it is to detect embedding. As such, this work builds upon a previously funded software program, ANNTS (Artificial Neural Network for Wavelet Steganalysis), which simply scanned an image for hidden content.

METHODOLOGY

The objective of this project is to develop a platform independent software package to perform blind steganalysis for the detection of stenography used in child pornography.

Specific project goals are:

- Expand the capabilities of ANNTS and enable the software to inspect multiple images. The current capability of ANNTS is to scan a single image. By modifying the code, a file folder of image data can be inspected and the results written to a file. Similarly, by modifying the software user interface, the meaning of labels written on screen can become clearer.
- Add two more targeted detection techniques that use known ebbed signatures to detect payload information. ANNTS currently utilizes four, and this project adds two more to scan for jpeg embedded images. The new techniques are selected based on their potential use for illegal criminal activity and interest by crime laboratory forensic scientists.
- Develop a “blind” detection technique that uses general image statistical measures to detect payloads. These general techniques will be based on the *dual-tree complex wavelet transform* (DC-CWT) and a *partially ordered Markov model* (POMM) for image data.
- Add detection schemes to the Graphical User Interface (GUI) of ANNTS, and rewrite the software code in JAVA to make the steganalyzer useful and available in a cross-platform package to its users.

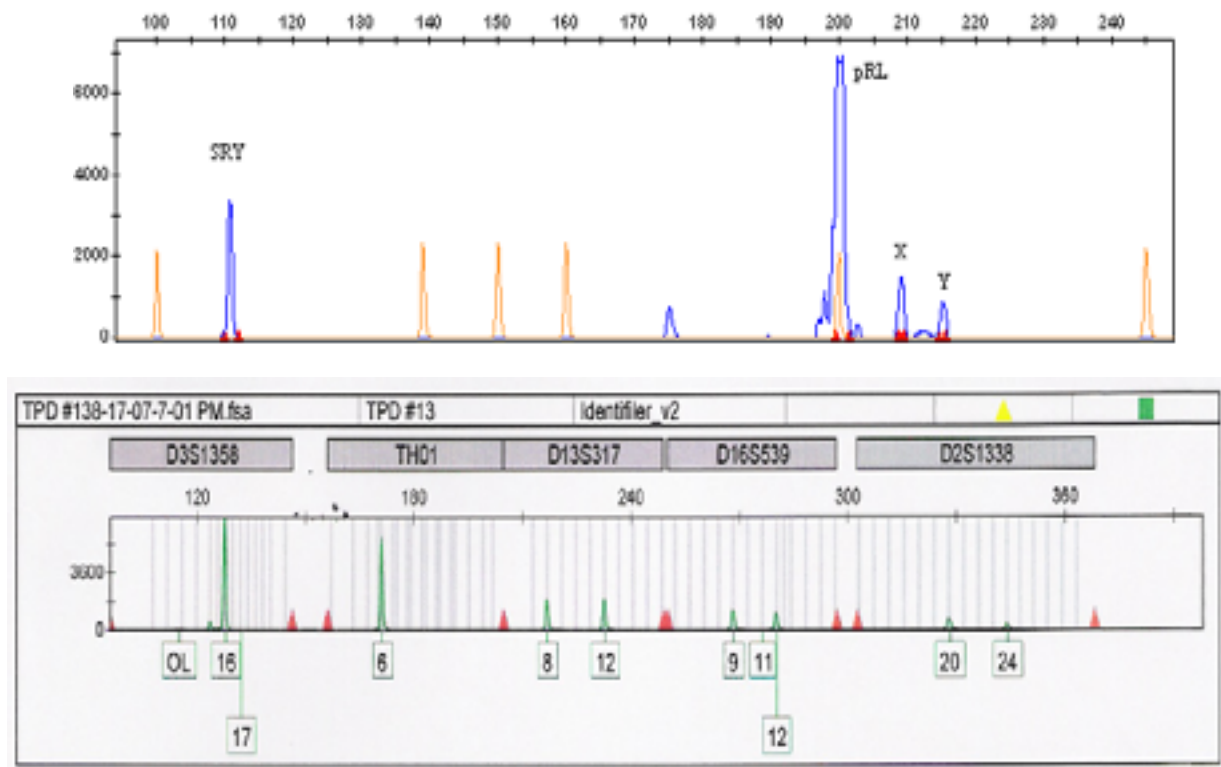


Figure 1. DNA sample exhibiting characteristics of partial degradation: Q-TAT and Identifiler results (green loci only).

ACCOMPLISHMENTS AND ONGOING WORK

A program called Canvass was developed to perform blind steganalysis on JPEG images. JPEG image steganography was targeted as JPEG images can easily be changed to remove “redundant” parts of a visual scene and replace them by “important” visual information represented by special JPEG format. Also, JPEG works in the transform domain of the Discrete Cosine Transform.

Two stenographic methods were added to scan for JPEG embedded images. They were: S-tools and JPHide & Seek. Both were chosen due to their potential use for illegal criminal activity and interest by crime laboratory forensic scientists. S-tools was later deleted due to its impractical nature of submitting batch jobs as it currently can only be run on a single image. Subsequently, five different stenographic methods were analyzed: JSteg, JPHide & Seek, OutGuess, F5 and StegHide.

The back end of Canvass uses a stochastic model called partially ordered Markov models (POMMs) to model (quantized) discrete cosine transform coefficients. The POMMs model produced 98 statistical features used in a support vector pattern detector. Mahalanobis distance, univariate and multivariate distances were used to determine the rank of image data feature sets of data from images. These distance measure tools are useful for comparing the similarity of unknown data sets with known data sets. A POMM model was developed for the steganalyzer, incorporating it into the software code, and rewriting the software code in JAVA to make it accessible to forensic scientists.

TECHNOLOGY BENEFITS

The research provides crime laboratories with a software package that addresses basic steganography needs for forensic applications. Currently, few crime laboratories have the capability to scan web pages and folders on a computer system for hidden content.

COLLABORATION

This project is a collaborative effort between Iowa State University and the Iowa Division of Criminal Investigation (DCI). DCI serves as a consultant to the project and as a software tester and evaluator.

DISSEMINATION

Research results were presented at various national and international meetings and conferences. A manuscript on the research and its findings was prepared for submission to the Journal of Digital Forensic Practice. A technical report on the project and its findings was posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

- Davidson, J., Jalan, J. “Feature Selection for Steganalysis Using the Mahalanobis Distance.” Presentation made at the SPIE-International Society for Optics and Photonics, Media Forensics and Security XII Conference, January 2010, San Jose, CA.
- Davidson, J., Jalan, J. “Canvass: Steganalysis Forensic Software for JPEG Images.” Presentation made at Midwest Forensic Resource Center Annual Meeting, May, 2010, Mackinaw Island, MI.
- Jalan, J. “Canvass: A Software Tool for Forensic Steganalysis of JPEG Images.” Presentation made at the ADFSL Conference on Digital Forensics, Security and Law, May 2010, St. Paul, MN.
- Davidson, J. Jalan, J. “Steganalysis using Partially Ordered Markov Models.” Presentation made at the 12th Information Hiding Conference, June 2010, Calgary, Canada.

IMPLEMENTATION

A beta version of the software package was installed on a computer at the DCI laboratory. Staff received instruction for use, a user’s manual was also prepared.

Other potential users, or stakeholders interested in testing the CANVAS software are encouraged to contact the PI. Feedback will be used to make adjustments to the program.

The final software program will be demonstrated at workshops and conferences and will be made available to interested parties through the MFRC.

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How to detect hidden data

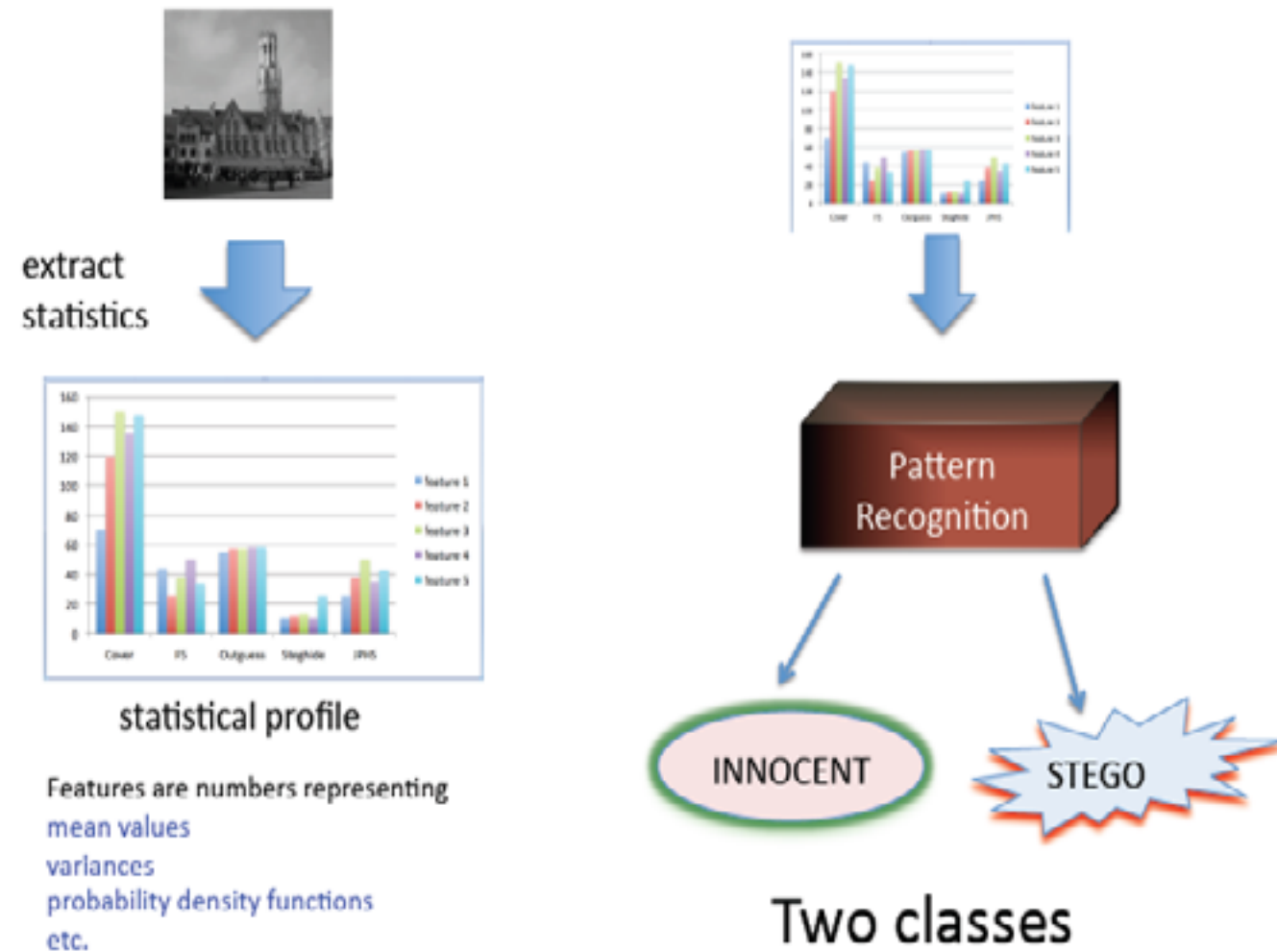


Figure 2. Flow chart of the steganalysis process

A Method for Lifting Bloody Impressions Using a Lifting Strip Containing Titanium Dioxide

FORENSIC TECHNOLOGY NEED

Bloody impressions are of great importance to the forensic science community as they are frequently encountered at crime scenes and on evidence submitted to the laboratory. Currently, there are a variety of techniques available for enhancing bloody fingerprints on non-porous, semi-porous and porous surfaces. Yet, depending on the surface porosity and background color, they require different enhancement processes.

In addition, many of the processes require the use of toxic chemical reagents and need to be conducted in a laboratory setting. Since impression evidence in blood cannot always be removed from the crime scene for analysis in a laboratory setting, many potentially identifiable impressions can only be photographed and not enhanced.

TECHNOLOGY DESCRIPTION

In an earlier study, a lifting strip containing titanium dioxide was successful in lifting and enhancing bloody fingerprints from several non-porous and semi-porous surfaces of contrasting colors. The lifting strips worked best when activated with a surfactant. After drying, the strip could easily be removed from the surface, lifting the bloody fingerprint onto a contrasting white background for examination.

To improve upon the methods used in the initial research, the primary objective of this study is to develop a better quality lifting strip, and to optimize the lifting process.

METHODOLOGY

To achieve these objectives, specific goals of the project are to:

- Manufacture larger lifting strips conducive to lifting better handprints, footprints, and footwear impressions.
- Determine the effectiveness of the lifting strip on a larger variety of porous, semi-porous, and non-porous surfaces
- Conduct research trials lifting bloody handprints, footprints, and footwear impressions from different substrates
- Determine the saturation limits regarding the amount of blood lifted with each lifting strip
- Determine the effectiveness of the lifting strip on aged impressions
- Conduct trials of the lifted bloody impressions utilizing various enhancement techniques
- Determine the effectiveness of the lifts when lifting pure proteinaceous blood samples
- Conduct presumptive DNA testing on the lifted impressions to determine if DNA is destroyed during the lifting process

ACCOMPLISHMENTS AND ONGOING WORK

Substrate materials were collected and sized for the deposition of bloody fingerprints, thumbprints, handprints, footprints, and footwear impressions. Selected surfaces were treated with sealant, paint, and stain to provide a realistic view of what is encountered at actual crime scenes. Substrates were also categorized (non-porous, semi-porous, and porous) and cut into small (1.5" X 2") and large (8" X 8") sample pieces. Seventy-two research boards were diagrammed to secure substrate samples and lift impressions for the research trials.

Improved lifting strips were designed and manufactured for use in lifting bloody impressions and to address the needs of the forensic community. The strip was made by bonding various titanium dioxide mixtures containing Kodak Photo-Flo, Liqui-Nox detergent, 5-Sulfosalicylic acid, acetic acid, methanol, ethanol, various polymers, silica, and water. The membrane was saturated in the titanium dioxide mixture to allow a uniform and thorough covering of the mixture onto the membrane, and then dried to complete the bonding process. The saturation limit of the lifting strip was determined to be 65 μ L per square inch.

Bloody impressions were deposited on seven non-porous, eleven semi-porous, and four porous substrates with each substrate having a total of forty impressions. Nineteen of the 22 substrates produced lifts with individualizing ridge detail; including overall impression patterns, ridge paths, deviations and dimensional attributes. Only three substrates posed impression analysis challenges.

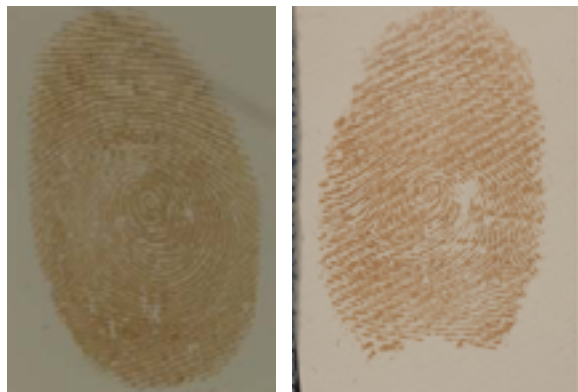


Figure 1. Lifted stained wood (left) and denim (right) fingerprint impressions.

Black vinyl had a rough textured background which muted some of the individualizing ridge detail in the lifted impressions. Yet, in the majority of the lifts, the overall impression patterns were still visible. In the denim, the dark background and prominent weave pattern made it difficult to determine the print quality while setting the deposition parameters, while on the canvas, the

displayed bloody weave patterns replicated the closely bound pattern of the fabric.

When coupled with proteins, the lifting strips were found to be inherently fluorescent under alternative lighting. The fluorescence lasted throughout the six month trial period with no diminishing intensity. Lifted impressions were consistent in contrast and fluorescent intensity without background interference in all the trials. The orange or yellow impressions were visualized on the dark colored light absorbent background of the lifting strips, creating an optimal quality brightly-fluorescing, high-contrast impressions for analysis.

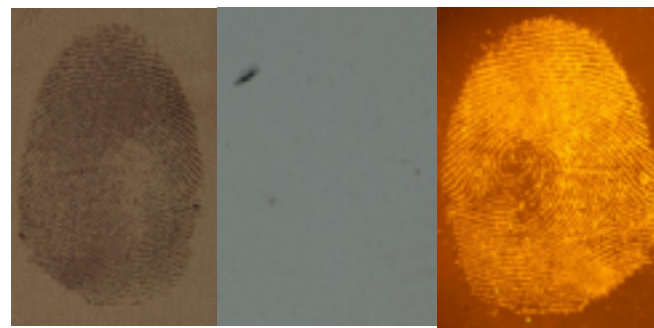


Figure 2. Cotton aged 6 months impressions: substrate (left), lifted (middle), and ALS lifted (right).

Extrinsic fluorogenic enhancement methods are not necessary or recommended for this method. Leucocrystal Violet (LCV), a fluorogenic reagent used to increase sensitivity and improve contrast when visualized with alternative lighting, greatly diminished the inherent fluorescence of bloody impressions on the lifting strip.

To determine the strip's effectiveness on larger lifts, bloody handprints, footprints, and footwear impressions were deposited on various substrates of differing porosity, ranging from sealed tile, linoleum, and stained wood to painted drywall, denim, and glass. The results were exceptional with all of the larger sized impressions displaying impression details with exceptional clarity and contrast.

To determine if DNA is destroyed during the lifting process, presumptive DNA testing on lifted impressions was conducted. Samples were run and are currently being analyzed. Findings are available, they are posted on the MFRC website.



Figure 3. Lifted impression of a painted drywall handprint.

TECHNOLOGY BENEFITS

Immovable objects from crime scenes cannot be brought back to the laboratory for analysis. Because of this, many potentially identifiable impressions may only be photographed and not enhanced thereby complicating the latent print identification process. With the use of the new uniform lifting strip, these problems can be alleviated while still providing optimal quality impressions for analysis.

The lifting strips are highly sensitive and inherently fluorescent when coupled with proteinaceous materials and excited with alternative lighting. The resulting fluorescent impressions are often greatly enhanced with exceptional clarity and contrast, even when the impressions were aged and faint or invisible under normal lighting conditions.

COLLABORATION

This is a collaborative effort between the Northville Police Department, the Michigan State Police

(MSP) Northville Forensic Science Laboratory (NFSL), and Madonna University. The MSP NFSL provides consultation and use of the photography equipment, while Madonna University provides laboratory space and financial oversight.

DISSEMINATION

The results of this study have been disseminated through presentations at forensic science conferences and at trade shows for the law enforcement community. A manuscript on the research findings was accepted for publication in the *Journal of Forensic Identification* and a technical report on the research and its findings was posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

- Zarate, J. "A Fluorogenic Method for Lifting, Enhancing, and Preserving Bloody Impressions." Presentation made at the Michigan State Police Annual Latent Print Unit Meeting, May 2010. Troy, MI.
- Zarate, J. "A Fluorogenic Method for Lifting Bloody Impressions Using a Lifting Strip Containing Titanium Dioxide." Presentation made at the Midwest Forensics Resource Center Annual Meeting, May 2010. Mackinac Island, MI.
- Zarate, J. "A Fluorogenic Method for Lifting, Enhancing, and Preserving Bloody Impressions." Poster presentation at the International Association for Identification (IAI) Annual Educational Conference, July 2010. Spokane, WA.
- Zarate, J. "A Fluorogenic Method for Lifting, Enhancing, and Preserving Bloody Impressions." Presentation made at the Midwestern Association of Forensic Scientists Annual Meeting, October 2010. Kansas City, KS.

- Zarate, J. “A Fluorogenic Method for Lifting, Enhancing, and Preserving Bloody Impressions.” Manuscript accepted for publication in the *Journal of Forensic Identification*, 2011.

IMPLEMENTATION

Requests for product samples were received from individuals and organizations worldwide and samples of fluorogenic blood lifting strips were distributed to forensic experts in the United States and Canada, and to forensic laboratories in Korea, France, Australia, and the United Kingdom.

In addition, validation studies have been conducted by the Forensics Laboratory of Kansas City, Kansas, to incorporate the method into their Policy and Procedures Manual. The Michigan State Police is currently using the method in their case work, including a recent homicide case.

Recently, the method has been described in a special section on detecting latent evidence in a textbook for law enforcement professionals (“Crime and Measurement: Methods in Forensic Science”, Myriam Nafte, and Brian Dalrymple, Carolina Academic Press, 2010).

Tri-Tech Forensics, Inc. (Southport, North Carolina) obtained the license for the lifting strips. Strips are commercially available under the name Zar-Pro Fluorescent Blood Lifting Strips and are marketed to law enforcement/crime scene investigators nationwide. The company is also marketing and distributing the product internationally.

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Application of Face Recognition Technology to Microstamped Cartridge Cases

FORENSIC TECHNOLOGY NEED

The National Research Council, in its 2008 published “Ballistic Imaging Report”, states that the fundamental assumption underlying forensic firearm identification (i.e. that every gun leaves unique microscopic marks on bullets and cartridge cases) has not been fully demonstrated scientifically.

Nevertheless, the council recommends that “microstamping” be researched to relate guns to fired ammunition, and for tracking the sources of illegally trafficked firearms. While the method shows promise, many questions remain, chief of which is the durability of the mark.

TECHNOLOGY DESCRIPTION

Microstamping is a technique used to etch individual serial numbers onto critical parts of a firearm. When the weapon is fired, the etchings are transferred to the cartridge case, thus allowing the cartridge cases to be related to the fired weapon.



Figure 1. 40 caliber Smith and Weston cartridge case imprinted with the firing pin.

Microstamping identification has been suggested as a way of providing an objective means to relate guns to fired ammunition. While the method shows promise, many questions remain, chief of which is the durability of the mark.

METHODOLOGY

The primary objectives of this project are to determine the ability of advanced imaging and recognition software to discern microstamped identifiers; the durability of microstamped identifiers on three handguns; and whether a microstamped identifier can be recovered using metallurgical etching techniques.

To achieve these objectives, the following steps are taken:

- Three different quality firearms are obtained, fired, cleaned, and polished at regular intervals
- The sequence of firing and cleaning is repeated until 1000 shots have been fired or the microstamped identifier is no longer available
- Cartridge cases are collected after every 100th and 101st sequence of shots and imaged using a surface optical profilometer
- Characterization of the identifier is carried out using an optical profilometer
- Metallurgical etching methods are used to determine which identifier can be removed from the surface of the firing pin

ACCOMPLISHMENTS AND ONGOING WORK

Three 9 mm semiautomatic handguns were acquired: a High Point (low quality), a Taurus

(moderate quality), and a Sig Sauer (high quality), along with the 12 additional firing pins for each handgun and the necessary ammunition for the study. The handguns and firing pins were sent to Pivotal Development LLC to place unique identifiers on the firing pins of the chosen weapons. The identifiers consisted of markings containing a series of both course and fine identifiers.



Figure 2. Example of a handgun to be used in the firing tests.

All of the ammunition to be used in the study was marked with sequential numbers, so that each individual cartridge can be identified in the sequence in which it will be fired. Currently investigators are awaiting shipment of the handguns and firing pins to begin the series of test fires, imaging of cartridge cases and firing pins, characterization/quantification of data, and removal/recovery of identifiers.

TECHNOLOGY BENEFITS

California signed into law a bill requiring microstamping that went into effect January 1, 2010. Other states are also investigating use of the technology. The findings of this study may help states determine if microstamping is indeed a viable technology to link a firearm to a cartridge. The results may also aid in firearm investigations by providing a quantitative measure of the degree of match.

COLLABORATION

This project is a collaborative effort among Ames Lab, Iowa State University, Jim Kreiser (a retired toolmark examiner for the State of Illinois), and the Microstamping Technology Center, a division of Pivotal development, LLC of Londonderry, NH.

Mr. Kreiser assists in obtaining the required firearms and ammunition and oversees the test firings and cleaning of the weapons. The Microstamping Technology Center marks the firing pins of the study weapons using the latest methods under development at Pivotal Development, LLC.

DISSEMINATION

Research findings will be disseminated through presentations at the Association of Firearm and Toolmark Examiners (AFTE) conferences and training seminars, as well as U.S. Department of Justice sponsored Firearms training and informational events. Presentations will also be made at the joint meeting of the Metallurgical Society/ASM International. A manuscript will be submitted to the Journal of Forensic Sciences, the AFTE journal, and other relevant peer-reviewed journals. A final technical report on the project will be posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

This is a new project with no presentations or publications to date.

IMPLEMENTATION

The findings of this project can have significant impact on the implementation of microstamping as a technique for firearm identification. Should the technology be mandated by legislatures, it is anticipated that wide-scale implementation will be less costly than the current National Integrated Ballistics Information Network (NIBIN) system.

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Discrimination of Dyed Cotton Fibers Based on UV-Visible Microspectrophotometry and Multivariate Statistical Analysis

FORENSIC TECHNOLOGY NEED

UV-visible (Ultra violet) microspectrophotometry (UV-MSP) is a popular technique used by trace evidence examiners for characterizing fiber color. While fast and generally considered to be highly discriminating, sample association with UV-MSP can be problematic due to sample heterogeneity and a lack of quantitative criteria for comparing spectra. A study is undertaken to establish the validity of UV-MSP for fiber examinations using multivariate statistical analysis techniques.

TECHNOLOGY DESCRIPTION

UV-visible microspectrophotometers are instruments designed to measure the UV-visible spectra of microscopic samples. With specialized software, UV-MSP's can be used to measure thin film thickness colorimetry and more. Preliminary results demonstrate that replicate spectra from fibers treated with different dyes form distinct clusters with spectra showing systematic differences. Statistical analysis further reveals that classes of dyes can be identified, leading to possible development of a dye classification scheme.

METHODOLOGY

The objectives of this project are to acquire a collection of dyed cotton fibers, acquire spectra from all samples using UV-MSP, and perform multivariate statistical analysis on the resultant spectra at two different laboratories to test the null hypothesis that the mean of a group of spectra from one laboratory is significantly different from the mean of a group of spectra from another laboratory. An additional test of consistency among data sets is employed by comparing the classification accuracy upon merging the two data sets to the classification performance of the separate tests.

Two popular data treatments are evaluated for the collection of spectra. The first is a calculation of first derivatives and the second is a transformation of the spectra into chromaticity coordinates. In each case, the multivariate calculations are repeated on the modified spectra and any differences in classification are noted to determine the extent to which spectra can be differentiated and shared between laboratories.

ACCOMPLISHMENTS AND ONGOING WORK

A set of twelve exemplars of various red dyed cotton fibers was acquired from a commercial source (Test Fabrics, Inc.) and from Stephen Morgan (University of South Carolina). Following standard MSP protocols, ten fibers were removed from each example and prepared using a technique modeled after that of the Indiana State Police Laboratory. Each of the exemplars was analyzed at IUPUI and the Indiana State Police.

A	Style 419, Bleached Mercerized Cotton Blend cloth dyed with Direct Red C-380	
B	Style 460, Bleached Cotton Interlock dyed with Fiber Reactive Red 120	
C	Style 460, Bleached Cotton Interlock dyed with Fiber Reactive Red 123	
D	STC EMPA 475 dyed with Reactive Red 195	
E	STC EMPA 495 dyed with Reactive Red 2	
F	STC EMPA 498 dyed with Reactive Red 228	

Figure 1. Dyed Cotton Exemplars from Testfabrics, Inc.

Three different data sets were created: First, Test Fabrics, Inc. fibers were analyzed by two individuals. Second, new fibers were selected from the twelve exemplars and analyzed at the Indiana State Police (ISP) Laboratory. Third, new fibers were selected from the same twelve exemplars and analyzed at IUPUI for inter-laboratory comparisons.

All data sets consisted of the truncated wavelength range of 350-800 nm to ensure that the data consisted of the same wavelength ranges. Samples were taken as absorbance values with ten scans taken at different locations for each fiber sample. Scans were then averaged to be used with Agglomerative Hierarchical Clustering (AHC), with full absorption data used in the remaining data analyses.

Statistical evaluation on the data was performed using Microsoft Excel XLSTAT 2009. The AHC dendrogram for fibers A - F (analyzed at IUPUI) showed the formation of three distinct classes based on the truncation line. Bifurcations to the right of the truncation line were more significant in determining the number of classes. In addition, the averages for each of the fiber dyes were used for AHC. Three classes were chosen to remain consistent with all other data sets because in the ISP data set, when more than three classes were used, replicates of some samples began to be split amongst the classes.

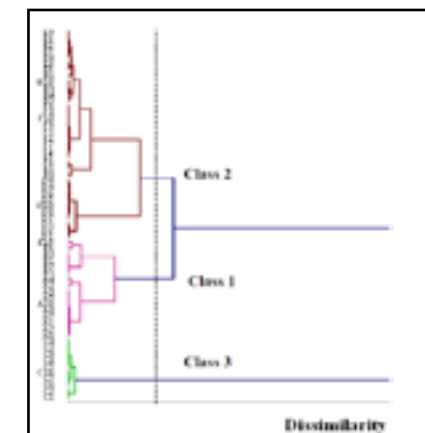


Figure 2. Dendrogram from AHC of Fibers A - F analyzed at IUPUI.

Principal Component Analysis (PCA) of all scans (rather than averages) indicated that 66.57% of the total variance in the data set can be explained by the first two principal components. The same three classes from AHC were color-coded to be distinguishable. Though the three classes are distinct, some overlap is present. Similar to AHC, Class 3 is more distinct than the other two classes.

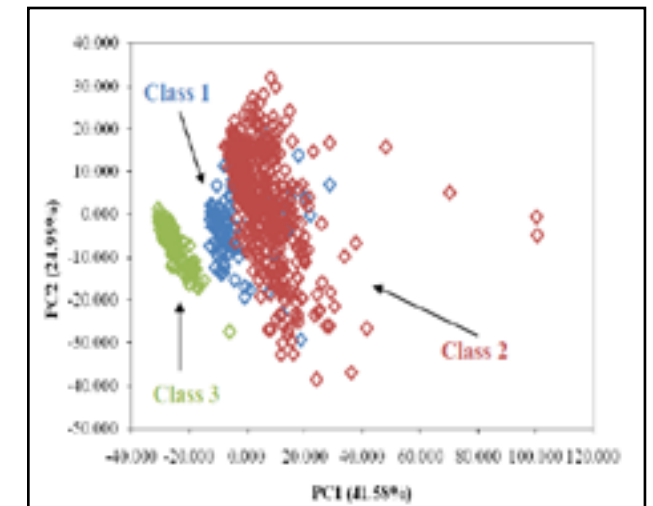


Figure 3. Observations plot from PCA.

Discriminant Analysis (DA) was performed using the three distinct groupings to be consistent with AHC. Minimal overlap can be seen between the three groups. Samples located along the diagonal in green represent those that were correctly classified, while samples in bold red outside of this diagonal are incorrect classifications. Overall, 99.17% of the samples were correctly classified.

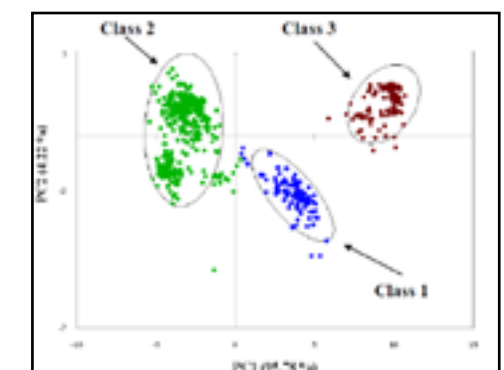


Figure 4. Observations plot from DA using 3 classes.

Current work is focusing on additional data analysis, in particular: identifying regions of the spectrum that are the most discriminating by calculating univariate Fisher ratios for every wavelength in a spectrum; performing comparisons of spectra obtained from instruments at IUPUI and the Indiana State Police; evaluating two popular data treatments for the collections of spectra to determine the extent to which spectra can be differentiated and shared between laboratories.

TECHNOLOGY BENEFITS

Trace evidence examiners generally do not have any quantitative basis for determining the probative value of an evidence type in question, which analytical method is the most discriminating for that evidence type, the effect of intra-sample variance on drawing their conclusion, or an objective criterion for comparing two samples. By applying multivariate statistical approaches to the UV-MSP data gathered from dyed cotton fibers, the understanding of trace evidence and the evaluation of the match of questioned and known samples will be greatly improved.

COLLABORATION

This project is a collaborative effort between Indiana-University-Purdue-University-Indianapolis (IUPUI), the Indiana State Police (ISP) Forensic Science Laboratory, and the University of South Carolina (USC). The ISP provides guidance to the project and participates directly in the analysis of cotton fibers using their CRAIG QDU 1000 instrument. USC provides cotton fibers as well as software specifically written for multivariate analysis of fiber spectra.

DISSEMINATION

The results of this research project have been disseminated at various chemical and forensic conferences, including the Fall Meeting of the Midwestern Association of Forensic Scientists and

at the annual meeting of the American Academy of Forensic Science. Manuscripts detailing the research and its findings will be submitted for publication in the *Journal of Forensic Sciences* and *Forensic Science International*. Upon completion of the project, a technical report will be posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

- Goodpaster, J. "Chemometric Analysis as a Means to Differentiate Class Evidence." Presentation made at the Federation of Analytical Chemistry and Spectroscopic Society (FACSS), October 2008, Reno, NV.
- Goodpaster, J. "Applications of Multivariate Statistics to Forensic Science." Presentation made at the Central Regional Meeting of the American Chemical Society (CERMACS), May 2009, Cleveland, OH.
- Goodpaster, J. "Analysis of Trace Evidence Using Microspectrophotometry and Multivariate Statistics." Presentation made at the NIJ Trace Evidence Symposium, August 2009, Clearwater Beach, FL.
- Morgan, S., Goodpaster, J., Lizeski, E. "The Use of Multivariate Statistics in Trace Evidence Investigations." Presentation made at the NIJ Trace Evidence Symposium, August 2009, Clearwater Beach, FL
- Goodpaster, J., Liszewski, E. "Forensic Analysis of Dyed Textile Fibers." *Anal. Bioanal. Chem.*, 2009, Vol. 394: 2009-2018.
- Goodpaster, J., Morgan, S., Liszewski, E. "Discrimination of Dyed Cotton Fibers Based on UV-Visible Microspectrophotometry and Multivariate Statistical Analysis." Presentation made at the Fall Meeting of the Midwestern Association of Forensic Scientists, October 2010, Kansas City, KS.

IMPLEMENTATION

Project findings and results are currently being evaluated to serve as guidelines for fiber examiners at the ISP Lab and potentially also the Scientific Working Group on Materials Analysis.

CONTACTS

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Informing the Judgments of Fingerprint Analysts Using Quality Metric and Statistical Assessment Tools

FORENSIC TECHNOLOGY NEED

The National Academy of Sciences (NAS), in its recent report *Strengthening Forensic Science in the United States: A Path Forward*, states that the latent print community has eschewed numerical scores and corresponding thresholds because those developed to date have been based on minutiae.

To address these concerns, a study is undertaken to determine if a “Quality” tool (to measure the clarity of the friction ridge) and a “Statistical Diagnostic” tool (to provide statistics that measure the strength of the evidence) might benefit fingerprint examiners during their decision making.

TECHNOLOGY DESCRIPTION

Specific objectives of the project are to:

- Determine how information regarding the clarity of friction ridge features (as provided in various formats to the participants) will inform the judgements of fingerprint analysts
- Determine how information regarding the strength of the corresponding friction ridge features (as provided in various formats to the participants) will inform the judgments of fingerprint examiners
- Determine how information regarding the strength of the corresponding features when provided by *other* fingerprint experts (“Expert Consultation”) will inform the judgments of participating examiners

METHODOLOGY

To achieve these objectives, the impact of the “Quality” and “Statistical Diagnostic” tools and

“Expert Consultation” on fingerprint examiner decision making is assessed. Six experimental groups are used, applying combinations of the three tools.

Variables	No LR tool	Expert Consultation	Diagnostic LR tool
No quality tool	Group 1 No quality tool No LR tool	Group 2 No quality tool Expert consultation	Group 2 No quality tool LR tool
Quality tool	Group 4 Quality tool No LR tool	Group 5 Quality tool Expert consultation	Group 6 Quality tool LR tool

Table 1. 2 X 3 Testing matrix to accomplish the stated objectives

The measured variables for the effect on examiner performance are the accuracy and reproducibility of the conclusions against the ground truth (including the impact on error rates) and the analyst variation during feature selection.

The hypothesis tested is that experts benefit from the implementation of tools that objectively assess clarity of friction ridge features and provide statistics regarding the strength of a match.

ACCOMPLISHMENTS AND ONGOING WORK

Approximately 600 latent print examiners were invited to participate in the study. Compact discs (CD-ROMs) were made and sent to each potential participant. The CD-ROM contained an applet that allowed the user to access a secure server at the Switzerland University of Lausanne (UNIL), each CD-ROM had a unique, random user name and password to maintain participant anonymity.

Approximately 200 examinees successfully logged in and participated in the study. A total of 176 participants completed all 12 trials resulting in 2112 trials to be analyzed. Of the 176 participants,

18 were trainees or technicians, leaving 158 latent print experts (who regularly perform case work).

Error rates, false discovery rates, and diagnostic test measurements (sensitivity, selectivity, rate of detection, etc.) were calculated for all participants (N=176). Error rates were reduced in groups 2 and 5, suggesting that natural collaboration and information exchange between fingerprint experts has benefit.

Given the difficulty of the cases, the false positive and false negative error rates were low. Trained experts (N=158) had lower false positive error rates, but higher false negative error rates, than novice trainees (N=13). The highest false positive error rates (15%) were observed in a small sub-set of experts (N=4) who did not annotate their ridge features of document the ACE process.

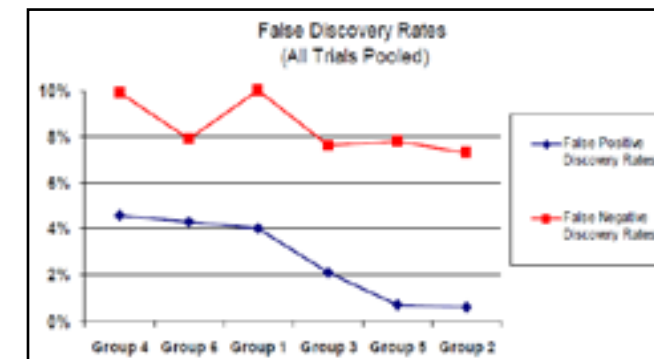


Figure 1. False discovery rates calculated by group.

The latent print image itself drove the mean number of and variance of minutiae selected. Experts using the Minutiae Consensus Map tool (group 2), or the Quality Map tool (groups 4 and 6), showed a higher accuracy and less variation than experts in the other groups with respect to which minutiae were annotated during the Analysis phase.

The tools did not appear to significantly influence the mean or variance of the number of minutiae annotated, but the group 2,4, and 6 tools did appear to reduce the number of false minutiae marked (i.e. the accuracy of the selected minutiae). Therefore,

a major benefit to the tools is that there is greater consensus and accuracy during the Analysis phase when selecting features.

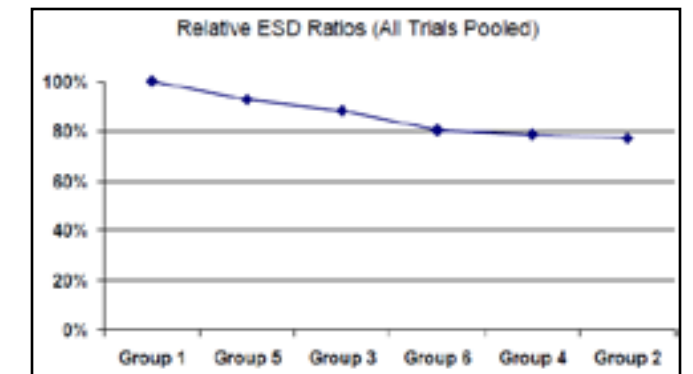


Figure 2. Relative ESD ratios showing a decreasing effect for groups using minutiae suggestion maps.

TECHNOLOGY BENEFITS

This research provides guidance on how best to integrate technology while maximizing analyst expertise and expert interpretation. In doing so, the research addresses the concerns of the NAS and provides an essential link between the theoretical/research models and actual examiner implementation during casework.

COLLABORATION

The project is a collaborative effort between the Minnesota Bureau of Criminal Apprehension (BCA), the University of Lausanne Switzerland (UNIL), and the Arizona Department of Public Safety Crime Laboratory. The principal investigator is a certified Latent Print Examiner at the BCA. He is also a Ph.D. candidate in the Forensic Science program at UNIL, and this project represents a portion of his work towards completion of his Ph.D. degree.

DISSEMINATION

The findings and results of this project will be presented at UNIL as part of his thesis defense and at national meetings. A manuscript of the research project and its findings will also be submitted to the

Journal of Forensic Sciences. Upon completion of the project, a technical report will be posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

- Langenburg, G., Neuman, C. “Moving Towards Using Statistics for Fingerprint Evidence in the Courtroom.” Presentation made at the 62nd Annual Meeting of the American Academy of Forensic Sciences, February 2010, Seattle, WA.
- Langenburg, G. “Informing the Judgments of Fingerprint Analysts Using Quality Metric and Statistical Assessment Tools.” Presentation made at the International Association for Identification (IAI) Annual Educational Conference, July 2010, Spokane, WA.
- Langenburg, G. “Uncertainty, Probability, and Statistics in Fingerprint Examination.” Presentation made at the NIJ Impression and Pattern Evidence Symposium, August 2010, Clearwater, FL.
- Langenburg, G. “Towards Greater Transparency in Documentation Tools for Communicating the Basis for your Conclusion.” Presentation made at the 2010 Canadian Identification Society 33rd Education Conference, September 2010, Orillia, Ontario, Canada.

IMPLEMENTATION

Depending on the outcome of the project, the results from the experiments may be used for training examiners on how to express statistical evidence in reports and testimonies. Recommendations may also be made to the latent print community via the IAI Standardization II committee and via SWGFAST (Scientific Working Group on Friction Ridge Analysis, Study, and Technology). These recommendations may include implementation of

“Quality” metric and “Statistical” measurement tools for assessing case work.

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Physical Matching in Trace Evidence: A Validation Study Using Automobile Parts

FORENSIC TECHNOLOGY NEED

A common scenario in Trace Evidence is a hit-and-run accident. Pieces of vehicular debris including glass, paint, plastic, wood, tape, and fabric are often left at the scene by a fleeing automobile. Crime scene investigators collect the evidence for physical matches to a suspect vehicle.

Physical matching is the parts to a suspect vehicle which are considered individualistic evidence. The assumption is that fracture matching is so intuitively obvious that a lay person could successfully execute the match. Yet, despite the wide use of the physical match, there is a lack of validation studies which document the reliability of physical matches by forensic examiners.

TECHNOLOGY DESCRIPTION

A physical match is a compelling piece of evidence used to place the vehicle at the scene of the accident. The primary objective of this study is to validate the physical matching process of questioned automotive parts to a known vehicle.

To test the hypothesis that relatively inexperienced persons can reliably determine the realignment of fragments, a study is undertaken to validate the physical matching process and to assess error rates associated with this type of examination.

METHODOLOGY

To achieve the objective, intact automobile parts from the front ends of vehicles commonly operated in the United States are obtained from salvage yards or from auto part outlet stores. The parts, in particular turn-signal housings, headlight assemblies, and side-mirror housings are fractured with a hammer to simulate an automobile accident.

Broken fragments of the assemblies are collected and re-assembled to:

- Demonstrate the physical matching process
- Document the success or failure of a group of laypersons to conduct physical matches
- Test the individuality of the physical match. More than 150 data points are collected and analyzed to calculate error rates

ACCOMPLISHMENTS AND ONGOING WORK

Ten different automobile parts were purchased from a local supplier, photographed and fractured to stimulate the damage done in a hit-and-run accident. Fracturing was achieved manually by clamping the part in a vise and striking it with a metal mallet wrapped in cotton cloth.

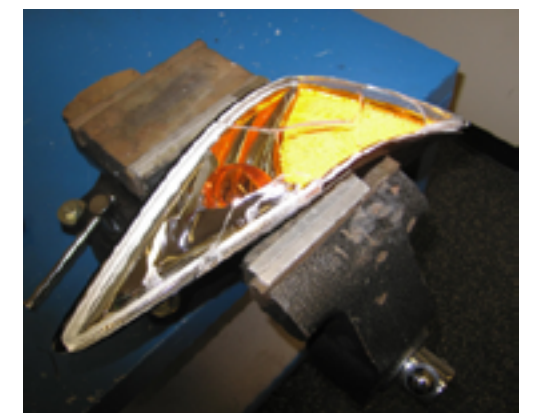


Figure 1. Assembly in vise prior to fracturing.

Ten students, serving as laypersons, volunteered to conduct the physical matches, five of which had taken fracture matching as a topic in a criminal justice course, but had no laboratory experience. The other five had no exposure to criminal justice. None were provided any background to fracture matching.

In Phase I, fragments from a single automobile assembly were collected and re-assembled by an experienced physical match examiner to determine physical matches. The results were documented and photographed.

In Phase II, a mixed collection of fragments from multiple broken automobile assemblies were matched to the proper assembly remnant. It was found that fracture simulation using the manual method does not fully mimic the fractures generated by an actual automobile impact.

Some of the fractures were less sharp and there was some bending that occurred with molded plastic. This caused some confusion with the students. Yet, it was decided that physical matches could still be validated and error rates calculated. A Type I error was defined as a failure to match at least one part, instead of several.



Figure 2. Assembly after fracturing.

It was found that in both Phase I and II, students completed the assemblies of fragments to remnants of the original much faster than anticipated. Data further showed that Type I (incorrect match) and Type II (rejected match) error rates were 0% in both studies.

These results seem to indicate that laypersons are capable to assemble fragments from an automobile lighting assembly back to the assembly remnant with 100% accuracy. This is the case even if more than one vehicle is involved in the accident.



Figure 3. Assembly with major fragments physically attached.

TECHNOLOGY BENEFITS

Physical and fracture matching have long been accepted in courts of law and have been considered intuitively obvious processes which even a layperson can understand and perform. In the process of defending a physical match on the witness stand, the analyst appeals to the layperson's concept of jigsaw puzzles and lock-and-key fits.

Relatively little has been done in the way of formal scientific processes to train or define physical matches relative to other fields of analysis. This study validates the principles of the criteria for physical matching and error rate calculation.

COLLABORATION

This project is a collaborative effort between the Trace Evidence Unit of the Wisconsin State Crime Laboratory Madison and the University of Wisconsin-Platteville (UWP) Department of Chemistry and Engineering Physics. UWP coordinates student employees and conducts statistical analyses of the research data.

DISSEMINATION

The results of this study are presented at annual meetings of the Midwestern Association of Forensic Scientists and the American Academy of Forensic Sciences. Publication in relevant peer-reviewed journals is also pursued. Upon completion of the

project, a technical report on the study and its findings will be posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

- Cornett, C., Wermeling, J. "Physical Matching in Trace Evidence: A Validation Study Using Automobile Parts." Presentation made at the Fall Meeting of the Midwestern Association of Forensic Scientists, October 2010, Kansas City, MO.

IMPLEMENTATION

It is anticipated that publication of the project findings in a peer-reviewed journal will provide a citation basis for laboratory findings used in court cases.

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Shape Measurement Tools in Impression Evidence: A Statistical Approach

FORENSIC TECHNOLOGY NEED

The National Academy of Science, in its 2009 report “Strengthening Forensic Science in the United States: A Path Forward”, notes the lack of statistical models in pattern evidence analysis. This project addresses this need by developing shape change analysis software that allows a multivariate statistical approach to explore the principle tenets of impression evidence: unique features of an object (acquired characteristics) and transfer of the unique features to a substrate.

TECHNOLOGY DESCRIPTION

In an earlier study, a shape analysis software was developed to examine principal causes of shape deformation in bitemarks as well as comparisons between dentations and bitemarks. This project builds upon that work by expanding the geometric morphometric software developed to investigate application in other areas of impression evidence, notably fingerprints and shoe impressions.

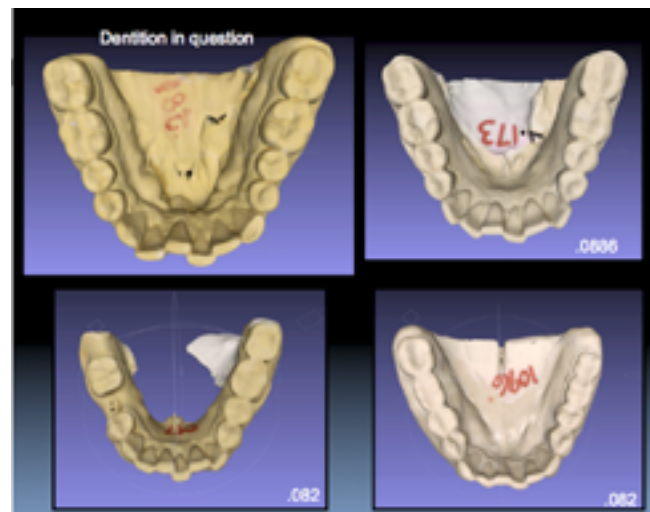


Figure 1. Shape analysis software using Procrustes distance to locate matches in database.

METHODOLOGY

The goal of this project is to explore the feasibility of applying shape measurement tools to fingerprint evidence and footwear impressions. By expanding the software used in the quantitative analysis of bitemarks, principal shape variation in fingerprint and footwear impressions can be investigated along with the repeatability and similarity of these types of pattern evidence. This will provide forensic practitioners with quantifiable validations of the quality of example items of pattern evidence.

To achieve this objective, the following procedures will be followed:

- Acquire impression specimens from the crime laboratory partner and also create experimentally as needed for the project
- Place landmarks on digital images of the specimens using TPSdig software and save coordinates of the landmarks in data files
- Visualize shape information by plotting landmark positions in superimposition
- Use IMP freeware to statistically analyze shape information
- Perform Canonical Variate Analysis (CVA) to determine relationships between groups of variables. Use CVA also to determine if shape information can distinguish between different categories of data
- Conduct Principal Component Analysis (PCA) to plot and visualize the principal variations of shape, allowing determination of which shape aspects are responsible for the most variation

- Determine Procrustes distances to measure closeness in shape. The statistic is used in the geometric morphometrics framework as a general-purpose measure of specimen similarity (a similarity metric)
- Determine parameters and limits of similarity and dissimilarity measurement error
- Determine measurement error rates using Root Mean Square, and Procrustes distances
- Determine inter- and intra-operator error, and match rates based on the effective resolution limit determined by repeated measures

ACCOMPLISHMENTS AND ONGOING WORK

This is a new project that has not yet started.

TECHNOLOGY BENEFITS

The research provides crime laboratories with a tool that addresses fundamental issues of pattern evidence: individuality and transferability. It also builds a framework for future statistical models to assign match probabilities based on population distributions.

COLLABORATION

This project is a collaborative effort between the State University of New York (SUNY) at Buffalo (SUNY-Buffero), Canisius College, and the Bureau of Criminal Apprehension (BCA) in St. Paul, Minnesota. Canisius College will develop the necessary shape measurement software, while the BCA will serve as a consultant to the project and perform software testing and evaluation.

DISSEMINATION

The results of this project will be presented at the American Academy of Forensic Sciences (AAFS) annual meeting. A manuscript on the project and its

findings will also be submitted to a peer reviewed volume or text. Finally, upon completion of the project, a final report will be posted on the MFRC website.

PUBLICATIONS AND PRESENTATIONS

This is a new project with no presentations or publications to date.

IMPLEMENTATION

The shape change analysis software developed for this project will be made available to crime laboratories via the Laboratory for Forensic Odontology Research (LFOR) website at SUNY-Buffero and through the Sheets' existing academic free-ware website at Canisius College. Links to access the software will be placed in the final report posted on the MFRC website. Workshops will be held at the AAFS annual meeting to provide education and training on the utility of the software.

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