



The author(s) shown below used Federal funding provided by the U.S. Department of Justice to prepare the following resource:

Document Title:	Sensitive Site Exploitation through Trace Chemical Analysis of Latent Fingerprints (SETCAF)
Author(s):	Daniel Pinto
Document Number:	251377
Date Received:	November 2017
Award Number:	2012-DN-BX-K038

This resource has not been published by the U.S. Department of Justice. This resource is being made publically available through the **Office of Justice Programs' National Criminal Justice Reference** Service.

Opinions or points of view expressed are those of the author(s) and do not necessarily reflect the official position or policies of the U.S. **Department of Justice.** 



1) ABSTRACT: In January 2013, we began a Basic Research project to develop sensitive site exploitation through trace chemical analysis of latent fingerprints (SETCAF). The project encompassed the following elements:

(I) Acquire latent prints from a diverse pool of 160 individuals, and in parallel establish a chemical print database to provide insight into fatty acid distribution in latent fingerprints from this donor pool. In addition, obtain a small sample of 15 to 20 smudges to determine if the same information regarding fatty acid distribution can be extracted from them. These fatty acid chemicals are intended to help determine personal characteristics, age, gender, and ethnicity that may be found in latent prints.

(II) Explore and execute hard (thermal/pyrolysis) and soft (supercritical carbon dioxide) extraction methods coupled with a broad spectrum chemical powder material for enhancing extraction of analytes from collected latent prints. The primary goal of this study was to provide a smart collection capability to gain enhanced information from these extracted chemicals, with a focus on fatty acids that would expand the role of the traditional print collection and analysis process into an additional repository of probative information.

(III) Look at the quantities of each fatty acid and their ratios from the diverse pool of individuals in order to compare age, gender, and ethnicity and note any differences to establish an initial chemical print database.

## 2) Executive Summary:

In January 2013, we began a Basic Research project to develop sensitive site exploitation through trace chemical analysis of latent fingerprints (SETCAF).

The primary goal of this project was to provide a smart collection capability to gain enhanced information from chemicals extracted from fingermarks and smudges, with a focus on fatty acids that would expand the role of the traditional latent fingerprint collection and analysis process into an additional repository of probative information. To achieve this goal, latent prints were acquired from a diverse pool of individuals and a chemical print database was established to provide insight into fatty acid distribution in latent fingerprints. Personal characteristics of interest were age, gender, and ethnicity. Identifying gender and ethnicity, among other demographics, from unknown fingerprints would be an invaluable investigative tool, and this study



suggests that further research in this area is certainly possible. Knowing a suspect's gender and/or ethnicity would allow investigators to narrow the pool of possible suspects, and help them to develop, analyze and prioritize investigative leads, by eliminating individuals as unlikely suspects.

## **Research design**:

Fingermarks were obtained by George Mason University (GMU). They were then analyzed both at GMU and at BAE Systems which is a subcontractor on this project. (See Appendix A).

Fingermark samples were obtained on standard 1X3" glass slides. The samples were labeled with an identifying number but no information about the donor was included to ensure anonymity. Samples were stored in closed containers at ~4 °C until analyzed. Fingermarks sent to BAE were to be dusted with BAE Systems fingerprint powder and lifted for analysis. However, numerous unsuccessful attempts were made to find a fingermark lifting tape that did not obscure the fatty acid signature or overwhelm the mass spectrometer detector. It was decided that the prints would be wiped from the slides using a moistened section of filter paper. The filter paper (Whatman grade QMA) was cut into small strips and folded in half to provide a sturdy wiping surface. The wipe was moistened with methanol to facilitate removing the oils from the glass. This methanol also served as the co-solvent which provided cleaner extraction and minimized carry-over in the system. It was determined that in most cases, a full fingerprint was too much sample for our system as the signal was saturated in the GC/MS and it would take several blank runs to clear down to baseline. Thus, a grid pattern was developed to position the slide so that a sample of approximately 1 cm<sup>2</sup> was measured to extract from the glass. At the operator's choice, greater or smaller sample areas were collected depending on the apparent volume of residue in the fingerprint. The paper was then loaded into the extraction chamber for analysis.

Several precautions were taken to minimize the risk of sample contamination. This is of extreme importance due to the ubiquity of the analytes found in the natural environment. All solvents and filter paper sheets were checked prior to use for contamination. All tools used to handle the samples were washed thoroughly with soap and water followed by a rinse using methanol. These items were checked periodically throughout the day to ensure integrity.

## **Methods:**

Analysis was performed on 160 samples by Thermal Desorption Unit coupled Gas Chromatography Tandem Mass Spectrometry (TDU-GCMS) and Supercritical Carbon Dioxide coupled Gas Chromatography Tandem Mass Spectrometry (SCCO2-GCMS), simultaneously. Although TDU-GCMS has the potential for chemical analysis of fatty acids from latent fingerprints, results from this chemical extraction



technique were not utilized due to poor reproducibility and extensive instrument troubleshooting necessary for consistent results. The alternative analysis by SCCO2-GCMS (Appendix A) demonstrated robust and consistent results, thereby, becoming the method of choice for the analysis of donor latent fingerprints.

Fatty acids ranging from C10 to C18, specifically decanoic acid, dodecanoic acid, tetradecanoic acid, pentadecanoic acid, hexadecanoic acid (palmitic acid), hexadecenoic (unsaturated), heptadecanoic acid, octadecanoic acid, and octadecenoic acid (unsaturated), were selected as analytes of interest for this experiment due to their ubiquitous expression in human epithelial cells and sweat glands, particularly by those present in hands, fingers, forehead and scalp. The literature and preliminary results indicated that the fatty acids selected are endogenously present in fingertips and transferred onto surfaces during contact (Ky, 2013; Pinto, 2014; Wiseman, 2014). It was observed that fatty acids can subsequently be extracted from latent fingerprints on a surface, in this case, a standard smooth 25 x 75 mm glass microscope slide, and chemically analyzed by SCCO2-GCMS (Pinto, 2014; Appendix A).

SCCO2-GCMS testing was performed on 160 latent fingerprints collected from donor participants on a microscope glass slide. Consistent elution peaks were observed and recorded for each successful analysis. Data was compiled into a master file which also included a calibration curve with known fatty acid concentrations from Sigma-Aldrich, allowing the conversion of raw peak height data into moles of fatty acid. Subsequently, normalization was done against hexadecanoic acid, a fatty acid normally present with a strong consistency in peak height and calculated moles (see Appendix A). A direct correlation was noted between hexadecanoic acid concentration and other coexisting fatty acids, C10 to C18, in multiple samples deposited by a single participant. Hexadecanoic acid presented cleaner chromatographic peaks with a typically high resolution when compared to the rest of the selected fatty acids (Figure 2.2.4.1, Appendix A). It was also observed that when a replicate latent fingerprint was tested, variations in the TIC count were directly proportional to the TIC count of the other species in the chromatogram (Appendix A). Therefore, hexadecanoic acid was initially chosen as an indicator for general fatty acid abundance (Figure 2.2.4.1, Appendix A).

The respective ratio of each fatty acid was calculated versus hexadecanoic acid, making hexadecanoic acid equal to 100% or 1.0, for each valid test. In scenarios where hexadecanoic acid was not detected, this was an indication of possible issues related to instrument error or inconsistencies in the specific sample being tested (Appendix A). Values of this nature were not included in the analysis of this study to minimize instrument related misrepresentation of a sample's fatty acid content.

The relative concentration of each fatty acid used for subsequent data analysis was obtained as a ratio of hexadecanoic acid (see Appendix A). This ratio was chosen to allow normalization of the data set. Normalized values to hexadecanoic acid typically ranged from 0.0 to 1.0, except for octadecanoic acid (C18) which had observed



relative concentrations >1.0. This means that the peak height and calculated moles were significantly higher than hexadecanoic acid. This observation also indicated that there could be some statistical power in utilizing octadecanoic acid as the base fatty acid for calculation of relative concentrations. Thus, it shed light onto the potential of octadecanoic acid adding some discriminating power when comparing fatty acids between individuals of different ages, genders and/or ethnicities.

After completion of SCCO2-GCMS analysis, data was compiled and merged with the metadata containing donor information specific to age, gender and ethnicity. Additional donor information, such as hand dexterity, contact with food, cosmetics or detergents, was also noted but not incorporated as a variable in the analysis. Initial statistical test results rejected the hypothesis that relative fatty acid concentrations were normally distributed within the utilized data set of 160 latent fingerprints. For this reason, non-parametric statistical techniques were considered most appropriate for further analysis, given that they make no assumptions about the underlying distribution of the data. Specifically, the Kruskal-Wallis H test, also known as the non-parametric one-way ANOVA on categories, was used to compare the abundance found for each fatty acid within categories of human physical characteristics such as age, gender and ethnicity.

# **Key Findings:**

## Fatty acids and Gender

Gender data analysis resulted in findings that seem to suggest that some fatty acids may be represented with a slightly varying concentration with respect to gender. The idea that gender may be determined by chemical compounds in fingerprints was previously speculated and tested yielding positive results in small sample sizes, but when sample size increased, no significant difference was noted (Asano et al. 2002). Other research suggests larger scale experiments on fatty acids as they are the predominant component of the superficial layer (Shetage et al. 2015). Out of the nine fatty acids studied, octadecanoic acid being used for ratio calculations, pentadecanoic acid was the most suggestive with a p-value 0.002 for the interaction between male and female results, yet lacking robustness. Detailed statistical analysis was not sufficient to make clear conclusions out of these results. However, with further research this is an area that might have some potential.

## Fatty acids and Ethnicity

Our study of ethnicity demonstrated that while most the fatty acids seemed to have no significant difference when comparing individuals of different ethnic groups (White Caucasian, African American and Hispanic), decanoic acid portrayed suggestive variations. Specifically, decanoic acid may be worth exploring for its potential to differentiate White Caucasians from Hispanic populations (p-value, 0.005), although robustness of results needs to be improved. Decanoic acid also showed interesting



results in the tests between White Caucasian and African American latent fingerprints (p-value, 0.028). From this data, we could conclude that the greatest difference in decanoic acid is observed between White Caucasian and African American populations, and that the Hispanic population decanoic acid concentration lies somewhere in between.

## **Conclusions:**

The goal of this project was to develop a method for analyzing the chemicals present in latent fingerprints that were transferred from the individual depositing them on a crime scene. Current fingerprinting practices are useful for the identification of latent fingerprints against a known fingerprint in a repository such as the Integrated Automated Fingerprint Identification System (IAFIS). However, latent fingerprints contain biochemical information with potential for forensic science applications. This project focused on investigating the potential of fatty acids, inherent in latent fingerprints, to be used to determine an individual's age, gender or ethnicity.

Identifying gender and ethnicity, among other demographics, from unknown fingerprints would be an invaluable investigative tool, and this study suggests that such identification may be possible with further research in this field. An increased number of samples Knowing a suspect's gender and/or ethnicity would allow investigators to narrow the pool of possible suspects, and help them to develop, analyze and prioritize investigative leads, by eliminating individuals as unlikely suspects.

Current forensic science practices consider DNA as the gold standard of forensic evidence. However, insufficient amounts of DNA in latent fingerprints and lack of a match in the Combined DNA Index System (CODIS) are limitations that SETCAF does not have. Fatty acids are ubiquitously present in fingers and transferred to latent fingerprints (Appendix A). In addition, some of the limitations of conventional fingerprinting, such as a fingerprint smudge, does not interfere with SETCAF. Further research of fatty acids in latent fingerprints and SETCAF present potential for determination of an individual's age, gender, and ethnicity.

To advance the application of SETCAF into the crime scene and to be able to make confident predictions of an individual's age, gender, and ethnicity, number of samples beyond the 160 from this study is necessary. We estimate that a number above 1000 samples will allow discerning an individual's age gender or ethnicity via fatty acids in latent fingerprints. This information will allow investigators to narrow their pool of suspects, apply additional forensic testing if warranted, and develop and analyze more discerning and relevant leads, which can lead to more expeditious resolution of cases that might otherwise languish for years.



Fingerprint analysis remains a viable forensic science and investigative tool. The comparison of known prints, contained in databases by the FBI, DHS, DOD, state and local governments to unknown fingerprints collected at a crime scene continues to be a core forensic science discipline. If useable fingerprints, containing enough fingerprint minutiae, are recovered at a crime scene, those prints are compared to known prints from the appropriate government fingerprint databases. If no identification is made of the unknown print because the offender's prints are not in the database, the investigative value of those prints diminishes significantly until a known suspect is identified. Currently, to our knowledge, the US Government does not apply a technique such as the one suggested by this study. SECAF could easily become a method used for identification, especially in cases where the suspects fingerprints are not in IAFIS. SETCAF would then provide law enforcement agencies with age, gender and ethnicity of a suspect and aid in the resolution of an investigation.

# 3) Performance: Significant Accomplishments

- Developed protocols to analyze the samples for fatty acid content and designed a method for collecting fingerprint residue on a glass slide, extracting the residue using chloroform, derivatizing it and then directly injecting it into the GCMS for detection.
- Devised a method of standardizing print collection conditions to include use of a scale to maintain consistent pressure of print application, humidors or similar containers to maintain temperature and humidity, and opaque containers to block out light, since it is known that light levels affect degradation rates.
- Obtained 160 IRB approved finger print samples for analysis.
- Analyzed samples using both the BAE extraction powder (at BAE) and a normal extraction process at (GMU).
- Evaluated the data by performing detailed statistical analysis and established a chemical matrix that provides info regarding age, gender, and ethnicity using a fatty acid extraction technique.

# 4) Technical Report

**ABSTRACT**: The primary goal of this project was to provide a smart collection capability to gain enhanced information from chemicals extracted from fingermarks and smudges, with a focus on fatty acids that would expand the role of the traditional print collection and analysis process into an additional repository of probative information. To achieve this, latent prints were acquired from a diverse pool of individuals and a chemical print database was established to provide insight into fatty acid distribution in latent fingerprints. Personal characteristics of interest were age, gender, and ethnicity.



## **RESULTS:**

SETCAF was performed on 160 samples by TDU-GCMS and SCCO2-GCMS, simultaneously. Although TDU-GCMS has the potential for chemical analysis of fatty acids from latent fingerprints, results from this chemical extraction technique were not utilized due to poor reproducibility and extensive instrument troubleshooting necessary for consistent results. The alternative extraction technique, SCCO2-GCMS (Appendix A), demonstrated robust and consistent results, thereby becoming the method of choice for the analysis of donor latent fingerprints.

Fatty acids ranging from C10 to C18, specifically decanoic acid, dodecanoic acid, tetradecanoic acid, pentadecanoic acid, hexadecanoic acid (palmitic acid), hexadecenoic (unsaturated), heptadecanoic acid, octadecanoic acid, and octadecenoic acid (unsaturated), were selected as analytes of interest for this experiment due to their ubiquitous expression in human epithelial cells and sweat glands, particularly by those present in hands, fingers, forehead and scalp (Shetage et al., 2014). Literature and preliminary results by Katherine Ky, James Wiseman, and Daniel Pinto indicated that fatty acids selected are endogenously present in fingertips and transferred onto surfaces during contact (Ky, 2013), (Pinto, 2014), (Wiseman, 2014). It was observed that fatty acids can subsequently be extracted from latent fingerprints on a surface, in this case a standard smooth 25 x 75 mm glass microscope slide, and chemically analyzed by SCCO2-GCMS (Pinto, 2014), (Appendix A).

SCCO2-GCMS testing was performed on 160 latent fingerprints deposited by donor participants on a microscope glass slide. Consistent elution peaks were observed and recorded for each successful analysis. Data was compiled into a master file which included a calibration curve with known concentrations and fatty acids from Sigma-Aldrich, allowing the conversion of raw peak height data into moles of fatty acid. Subsequently, normalization was done against hexadecanoic acid, a fatty acid normally present with a strong consistency in peak height and calculated moles (see Appendix A). A direct correlation was noted between hexadecanoic acid concentration and other coexisting fatty acids, C10 to C18, in multiple samples deposited by a single participant. Hexadecanoic acid presented cleaner chromatographic peaks with a typically high resolution when compared to the rest of the selected fatty acids (Figure 2.2.4.1, Appendix A). It was also observed that when a replicate latent fingerprint was tested, variations in the TIC count were directly proportional to the TIC count of the other species in the chromatogram (Appendix A). Therefore, hexadecanoic acid was initially chosen as an indicator for general fatty acid abundance (Figure 2.2.4.1, Appendix A).

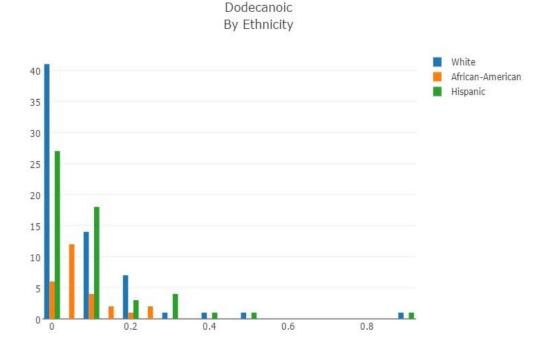
The respective ratio of each fatty acid was calculated versus hexadecanoic acid, making hexadecanoic acid equal to 100% or 1.0, for each valid test. In scenarios where hexadecanoic acid was not detected, this was an indication of possible issues



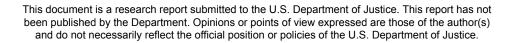
related to instrument error or inconsistencies the specific sample being tested (Appendix A). Values of this nature were not included in the analysis of this study to minimize instrument related misrepresentation of a sample's fatty acid content.

The relative concentration of each fatty acid used for subsequent data analysis was obtained as a ratio of hexadecanoic acid (see Appendix A). This ratio was chosen to allow normalization of the data set. Normalized values to hexadecanoic acid typically ranged from 0.0 to 1.0, with the exception of octadecanoic acid (C18) which yielded relative concentrations >1.0. This means that the peak height and calculated moles were significantly higher than hexadecanoic acid. This observation also indicated that there could be some statistical power in utilizing octadecanoic acid as the base fatty acid for calculation of relative concentrations. Thus, it shed light onto the potential of octadecanoic acid adding some discriminating power when comparing fatty acids between individuals of different ages, genders and/or ethnicities.

After completion of SCCO2-GCMS analysis, data was compiled and merged with the



**Figure 1** Histogram of relative concentrations of dodecanoic acid. Graph shows absence of normal distribution in data, as the greatest number of observations (y-axis) for relative concentrations (x-axis) as a function of hexadecanoic acid fall closer to 0.0 and only a few observations for higher relative concentration were metadata containing donor information specific to age, gender and ethnicity. Additional donor information, such as hand dexterity, contact with food, cosmetics or detergents, was also noted but not incorporated as a variable in the analysis. Initial statistical test results rejected the hypothesis that relative fatty acid concentrations





were normally distributed within the data set of 150+ latent fingerprints (**Figure 1**). For this reason, non-parametric statistical techniques were considered most appropriate for further analysis, given that they make no assumptions about the underlying distribution of the data. Specifically, the Kruskal-Wallis H test, also known as the non-parametric one-way ANOVA on categories, was used to compare the abundance found for each fatty acid within categories of human physical characteristics such as age, gender and ethnicity.

Data displayed on Figure 1 indicated that hexadecanoic acid might not be the best normalizing fatty acid for the data set. A series of computational analyses were performed to evaluate the suitability of each of the nine fatty acids to act as a normalizing agent for the remainder of the dataset. The analysis consisted of first calculating the ratio of each fatty acid to the fatty acid that was being evaluated. These relative concentrations were then grouped into metadata categories (i.e., age group, gender, and ethnicity) and tested for normality based on Jarque-Bera normality test. When used as a ratio denominator, octadecanoic acid had the least number of groups failing the normality test at a significance level of alpha = 0.1, and thus contributed the greatest normalizing effect on the data. The normalizing effect may be due to the relatively low dispersion of measured levels of octadecanoic acid in the sample of latent fingerprints. Table 1 below shows a variation analysis on raw data, prior to normalization.

Fatty Acid	Quartile Coefficient of	Coefficient of
	Dispersion	Variation
Decanoic	0.58	1.03
Pentadecanoic	0.76	1.34
Octadecanoic	0.69	1.34
Hexadecanoic	0.74	1.35
Tetradecanoic	0.71	1.49
Octadecenoic	0.82	1.84
Hexadecenoic	0.85	1.91
Heptadecanoic	0.79	2.74
Dodecanoic	0.52	3.01

**Table 1** Data analysis showing the variation observed on raw data set for SCCO2-GCMS prior to normalization. This table shows octadecanoic acid among the fatty acids with least variation.



As can be seen, octadecanoic acid falls among the least variable fatty acids in the population studied. Because of the relatively low variation and its normalizing effects, octadecanoic acid was chosen as the fatty acid to normalize the entire data set. It is also important to mention that all subsequent analyses and graphs were created excluding any outliers identified by Tukey's interquartile range method.

# TASK 1: Key fatty acids in the discrimination of Age in latent fingerprints

Participants who provided fingerprint samples to this study were categorized into five age groups: 0-7 years, 8-15 years, 16-22 years, 23-39 years, and 40+ years. For each fatty acid, relative concentrations within age groups were examined via pairwise Kruskal-Wallis tests to determine whether there were significant differences in the mean concentration of the acid. That is, for each fatty acid, we tested the null hypothesis that the mean level of the relative concentration is the same for "sample a" and "sample b" (sample "a" being a specific age group and sample "b" being an age group other than that represented by sample "a") The p-value of the test result represents the probability that the null hypothesis is true, and if p < 0.05 then the null hypothesis is rejected, implying there may be a statistically significant differences in all cases for fatty acids tested, but it did yield some indications of differences that may or may not be ubiquitous to age specific latent fingerprints. Table 2 below contains the most significant data analysis results for age.

Fatty Acid	Sample	Sample	Group 1	Group 2	p-value
	Size 1	Size 2			
Decanoic	53	34	23 To 39	16 To 22	0.000276265
Decanoic	32	34	40 And Up	16 To 22	0.001628905
Heptadecanoic	30	53	40 And Up	23 To 39	1.69475E-06
Heptadecanoic	33	53	16 To 22	23 To 39	9.50203E-05
Hexadecenoic	53	32	23 To 39	40 And Up	0.003245213
Octadecenoic	57	32	23 To 39	16 To 22	7.91498E-06
Octadecenoic	57	29	23 To 39	40 And Up	0.000400483
Pentadecanoic	32	34	40 And Up	16 To 22	0.010672555
Tetradecanoic	33	32	16 To 22	40 And Up	0.021659907

 Table 2. Key Fatty acids with potential for discrimination of age.

Observations revealed that the fatty acids mentioned in Table 2 might be most useful to discriminate between the following age groups: 16-22 and 23-39 years old; 16-22 and 40+ years old; and 23-39 and 40+ years old. Figure 2 below provides a visual example of the differing distributions of a fatty acid's relative concentration between age groups.

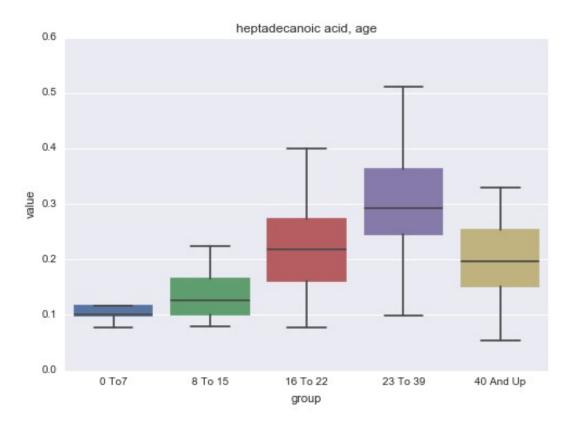


Figure 2. Heptadecanoic acid as a function octadecanoic acid, p-value 1.7E-06

## TASK 2: Key fatty acids in the discrimination of gender in latent fingerprints

Using an identical methodology to that described above, each fatty acid was tested for differences in mean relative concentration between participant's gender categories. Testing revealed three main fatty acids whose relative concentrations suggest underlying differences in fatty acid profile of latent fingerprints between male and female samples: hexadecenoic (unsaturated) acid, pentadecanoic acid and octadecenoic (unsaturated) acid (Table 3). The rest of the fatty acids tested did not reveal strong differences, indicating that their relative concentrations may not differ between males and females.

Fatty Acid	Sample Size 1	Sample Size 2	Group 1	Group 2	p-value
Pentadecanoic	62	67	Male	Female	0.002399073
Octadecenoic	57	71	Male	Female	0.003582072
(unsaturated)					
Hexadecenoic	61	70	Male	Female	0.030109983
(unsaturated)					

*Table 3.* Key Fatty acids with potential for gender discrimination. Tests were conducted after excluding outliers within the fatty acid groups.



Despite the interesting findings revealed by the Kruskal-Wallis test, further analysis of hexadecenoic acid, pentadecanoic acid and octadecenoic acid seems to indicate that these distinctions are weak or possibly spurious. Box plots presented in Figure 3 demonstrate that concentrations of pentadecanoic acid, whose test results were the most suggestive of the three fatty acids, may not be particularly useful for discriminating between genders.

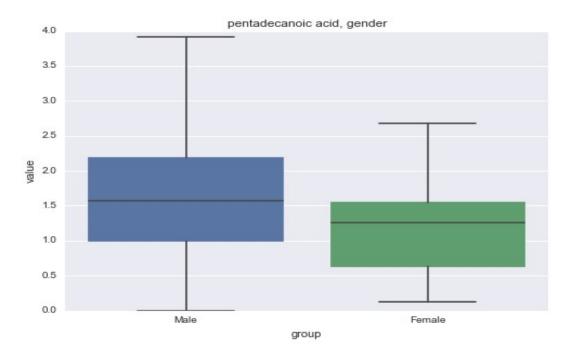


Figure 3. Pentadecanoic acid as a function of octadecanoic acid for gender, p-value 0.002.

Figure 3 represents male and female relative fatty acid concentrations and compares them showing some distinction at the mean values but also illustrates an overlap of data between both gender categories. Thus, despite a low p-value, the relative concentration of pentadecanoic acid is not sufficient to accurately discriminate between genders.

TASK 3: Ke	y fatty acids	in the disc	rimination	of Ethnicity	in latent fingerprints	

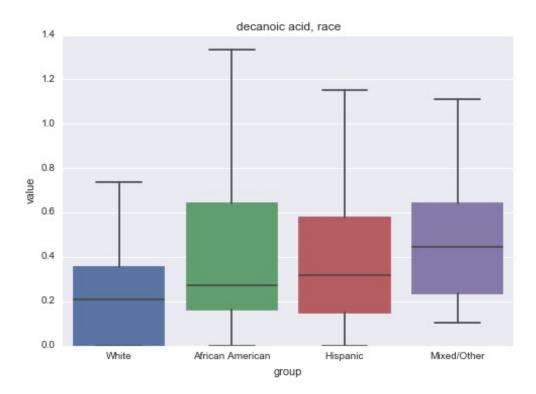
Fatty Acid	Sample	Sample	Group 1	Group 2	p-value
	Size 1	Size 2			
Decanoic	46	55	Hispanic	White Caucasian	0.005446603
Decanoic	55	24	White	African	0.027927073
			Caucasian	American	

Table 4. Key Fatty acids with potential for discrimination of ethnicity.



Data collected from participants was categorized into Hispanic, White Caucasian and African American ethnic groups. Like the analyses above, certain fatty acids were demonstrated to have slight potential for differentiating between participants' ethnicities, while others did not reveal strong differences. Table 4 demonstrates that the relative concentration of decanoic acid could be significantly different between Hispanic, African American, and White Caucasian samples in the data set, possibly indicating that the differences between fatty acid content in latent fingerprints vary per ethnic background.

Figure 4, below, portrays the distribution of mean values for decanoic acid. As it can be seen, there is a slight increase in mean value from White Caucasian to African American to Hispanic (Figure 4). The most noticeable p-values for these observations were 0.005 for White Caucasian versus Hispanic and 0.028 for White Caucasian versus African American. In both cases the null hypothesis was not supported. On the other hand, observed p-value for African American and Hispanic did not reveal any suggestive difference (p-value 0.931,  $n_H = 46$  and  $n_{AA} = 24$ ).



**Figure 4.** Box plot of ethnic groups studied for decanoic acid as a function of octadecanoic acid. Mixed/other races were not analyzed but data was collected. Outliers excluded. P-value 0.005 for White Caucasian versus Hispanic and 0.028 for White Caucasian versus African American.



# DISCUSSION

Latent fingerprints have been shown to contain information, other than traditional ridge patterns, relevant to crime scene investigation and in support of circumstantial evidence that may have the power to elucidate information such as the age, gender and ethnicity of an individual (Asano et al., 2002), (Frick et al., 2015), (Girod et al., 2012). The presence of chemical compounds, such as triglycerides, wax esters, free fatty acids, and squalene, in the analysis of fingerprints have been compounds of major focus in an effort to relate chemical composition to physical characterizations (Michalski et al., 2013). While the relative fatty acid concentrations discussed above are not sufficient to accurately discriminate between age, ethnicity, and gender categories, results indicate that some suggestive differences in fatty acid composition may exist between these groups. Hopefully, these differences will be further elucidated in the future by larger scale experiments (n > 1000).

In this experiment, a double blind chemical analysis method was designed to ensure anonymity of donor characteristics and protection of personally identifiable information. Donor information was collected only after approval by

GMU's Institutional Review Board (IRB) and NIJ's Human Subject Testing Officers, and donor consent. Each donor was assigned a number and names were always kept confidential. Metadata for each donor was immediately stored in a locked file cabinet behind locked doors in the Forensic Science Program Offices at GMU. A limited number of individuals had access to this information to prevent errors in data reconciliation. Sample collection was kept consistent by instructing the donors to rub the thumb and middle finger against their forehead for three seconds, for a total of 4 fingerprints. Donors were also asked not to wash their hands before collection of fingermarks. Even though a sample collection protocol was established to ensure minimal variation between collections, there are two main issues that were addressed. First, collection of multiple samples from a single individual presents the opportunity for loss of fatty acid material due to repeated contact with the glass slide. To prevent this, four samples were collected methodically from different fingers (thumb and middle finger from each hand). Second, the chance that each finger contains significantly different fatty acid concentration due to surface area was addressed by using a grid system to measure down to  $1 \text{ cm}^2$  (Appendix A).

Some accomplishments included the development and refining of a novel combination of instruments to perform SCCO2 extraction of latent fingerprints. BAE Systems has shown that the instrument has adequate sensitivity and extraction/transfer efficiency to perform chemical analysis on latent fingerprints (Appendix A). We have performed this analysis on approximately 160 samples to date with minimal instrument down time owing to the robustness of the system. In addition, one of the main issues originally encountered was the interference of the



adhesive compounds found in tape typically used in the collection of latent fingerprints from a crime scene. This issue was avoided by attempting a technique that consisted of attaching a G/F Whatman filter paper to the opposite side of the lifted print. This technique allowed retention of the adhesive compound while allowing fatty acids to adhere to the powder to migrate through the paper and be carried by the carrier gas (CO2) in the SCCO2-GCMS system. The analysis still yielded adhesive compound signals in the chromatogram, but when analyzed in single ion mode (SIM) peaks consistent with the fatty acids studied were clearly visible (Appendix A). Although this is a novel non-destructive approach to the analysis of latent fingerprints, that proves it possible to extract fatty acids from a latent fingerprint without destroying it, and with real potential for a direct application, further tests must be done to ensure the reproducibility of this specific nondestructive technique.

This experiment differs from other fingerprint composition studies in that it tries to accomplish the extraction and measurement of fatty acids from latent fingerprints, that is, fingerprints that are invisible to the unaided eye and may be collected by conventional CSI techniques with slight modifications in their tools (e.g. powder). Original work in this grant has shown that BAE System's powder works with comparable efficiency to conventional fingerprint powder. In the final stages of this experiment, fatty acids were extracted from dusted latent fingerprints without destroying the original fingerprints. This represents an important advance in the analysis of trace evidence for latent fingerprints. It not only allows collection of chemicals from latent fingerprints, but it also preserves evidence that may be used subsequently for additional testing.

## **TASK 1: Predicting Age:**

The age of an individual is directly related to the efficiency of metabolic processes naturally occurring in the body (Koenig et al. 2011). It is suspected that with increasing age the body loses its capability to efficiently synthesize compounds, such as fatty acids (Antoine et al. 2010). Therefore, this raises the question whether the variations in biosynthesis can be detected and utilized in forensic investigations. From data collected and analyzed in this study, the potential for discrimination is suggestive and further research is necessary to be able to use fatty acids in latent fingerprints to approximate a perpetrator's age. Some of the difficulties encountered that may have prevented this study from being successful at using fatty acids to approximate age are: 1) difficulty of collection. The difficulty of finding participants for this study from the age groups of 0 to 18 was directly related to their status of minors, adding an additional step since parental consent is necessary. While adults could give real time consent and donate latent fingerprints immediately, minors required prior consent by parents. Several minors interested in the study would not follow-up post parental consent, and special efforts, such as IRB approved announcements and planned collection sites, were necessary yielding only a few samples per attempt. Some of the scenarios observed were minors forgetting their consent form, not allowing them to participate in the study. 2) The collection and



recording of the exact age of the donor participant, as opposed to age group, would significantly increase the value of the data collected. Participants demonstrated certain skepticism towards donating their fingerprints. A crime scene investigation (CSI) effect was noticed when participants expressed concern that their fingerprints could easily be used to falsely incriminate them in a crime (Roane, 2005). When requesting voluntary information, such as exact age, the participants became more reluctant to donate their fingerprints. Participants were significantly more comfortable providing their age group rather than their exact age. Obtaining the exact age of the participant would allow a single variable analysis of the data of age as a function of time (in years) that may provide new insight into its correlation to fatty acid content in latent fingerprints and the difference due to aging.

## TASK 2: Fatty acids and Gender:

Gender data analysis resulted in findings that seem to suggest that some fatty acids may be represented with a slightly varying concentration with respect to gender. The idea that gender may be determined by chemical compounds in fingerprints was previously speculated and tested yielding positive results in small sample sizes, but when sample sizes increased, no significant difference was noted (Asano et al., 2002). Other research suggests larger scale experiments on fatty acids as they are the predominant component of the superficial layer (Shetage et al., 2015). Of the nine fatty acids studied, octadecanoic acid being used for ratio calculations, pentadecanoic acid was the most suggestive with a p-value 0.002 for the interaction between male and female results, yet lackied robustness. Statistics alone are insufficient to make a clear conclusion from these results. The understanding of fatty acids regarding biosynthesis, environmental factors, habits and diet may play an essential role in their presence in latent fingerprints. While there may be an underling genetic component to fatty acid differences in gender, observations were made that may contribute to the idea that certain habits, such as use of cosmetics, may contribute to the fatty acid profile of an individual. The answer may be in larger sample sizes that will either obscure the effect of outside factors or highlight their effect. Since fatty acids present in fingerprint residue have not been widely studied (Girod, A. et al. 2012), studies that look at the link between genetic regulation, fatty acid synthesis, and gender may assist in understanding the potential of fatty acids as a discriminatory tool.

It will also be necessary to identify what factor contributes the most to variations in concentration of fatty acids. For example, regulation of synthesis due to genetic factors, abundance/lack of certain dietary nutrients, or transmitted compounds from cosmetics and other commonly used products may contribute or suppress detection and abundance of fatty acids. Future research should focus on larger scale testing (n > 1000) and look at how outside factors contribute to the data. Further exploration of how environmental factors such as soap, food and cosmetics affect the fatty acid profile must be made to fully understand their role in fatty acids present in latent fingerprints.



To judge the discriminatory power of fatty acid concentrations in predicting gender, SETCAF data was examined via a Classification and Regression Trees algorithm, with 80% of the sample data chosen randomly to train the algorithm and the remaining 20% used to test the results. Results indicated as high as 75% accuracy in using the relative concentration of all fatty acids collected to predict gender; however, accuracy varied greatly based on different random splits of the data. These results were an indication that while multivariate analysis of all fatty acids may be useful in gender prediction, further exploration of the individual contribution of each fatty acid is necessary to find key fatty acids that directly impact our chances of making a prediction. We expect to find better discriminatory results for gender (as well as age and ethnicity) as the sample size increases above 1000 samples.

At this stage analysis of single fatty acids may yield insight into key differences in fatty acid contents among males and females. As seen in Figure 3 males seem to exhibit a higher relative concentration of pentadecanoic acid than females in general, but there is significant overlap in the distributions such that discriminating between categories based on this measurement alone would now be inconclusive. Nonetheless, the results suggest that the relative concentration of fatty acids may differ between males and females. Further studies may shed more light on the nature of these differences.

#### TASK 3: Fatty acids and Ethnicity:

Testing demonstrated that while most of the fatty acids seemed to have no significant difference when comparing individuals of different ethnic groups (White Caucasian, African American and Hispanic), decanoic acid had suggestive variations. Specifically, decanoic acid may be worth exploring for its potential to differentiate White Caucasians from Hispanic populations (p-value, 0.005), although robustness of results needs to be improved. Decanoic acid also showed interesting results in the tests between White Caucasian and African American latent fingerprints (p-value, (0.028). From this data, we could conclude that the greatest difference in decanoic acid is observed between White Caucasian and African American populations, and that Hispanic population decanoic acid concentration lies somewhere in between. Yet, as per the data, Hispanic population mean relative concertation overlaps too closely with African American. While we are observing mean values, and can make some observations, as seen in Figure 4, it is important to remember that overlap of concentration was observed for all three ethnic categories in this study. As sample size  $(n \ge 1000)$  increases it is suspected that the true results will become more apparent, whether the overlap among ethnic categories diminishes or it remains similar. For future research, decanoic acid may be a fatty acid worth focusing on.



# Conclusion

## Fingerprint analysis remains a viable forensic science investigative tool:

The comparison of known prints, contained in databases by the FBI, DHS, DOD, state and local governments to unknown prints collected at a crime scene continues to be a core forensic science discipline. If useable prints, containing enough fingerprint minutiae, are recovered at a crime scene, those prints are compared to known prints from the appropriate government fingerprint databases. If no identification is made of the unknown print because the offender's prints are not in the database, the investigative value of those prints diminishes significantly until a known suspect is identified. Currently, the US Government is not involved in further research and/or analysis of unknown prints to extract biological fluids from those prints to determine specific human markers such as donor gender or ethnicity or environmental factors such as indicators of drug and nicotine consumption.

The goal of this project was to develop a method for analyzing latent fingerprints that otherwise possess little evidentiary value because the quality of the latent print does not possess enough detail to identify the suspect. This project focused on future possible fingerprint examinations which would examine the fingerprint processing powder and test the biological fluid contained in the powder for personal and environmental components such as prescription medications, foods, cosmetics, nicotine, etc.

Identifying gender and ethnicity, among other demographics, from unknown fingerprints would be an invaluable investigative tool, and this study suggests that further research in this area is certainly possible. Knowing a suspect's gender and/or ethnicity would allow investigators to narrow the pool of possible suspects, and help them to develop, analyze and prioritize investigative leads, by immediately eliminating individuals as unlikely suspects.

DNA is clearly the gold standard of forensic evidence. However, from a crime scene perspective, the amount of DNA contained in the bodily fluids absorbed in the fingerprint powders are below the quantitative threshold for forensic DNA analysis. In addition, finger print smudges that are collected at a crime scene currently have no investigative value. If further research is conducted into bodily fluids extracted from the fingerprint powders as performed in this study, those once useless smudges can be used to extract relevant information such as the fingerprint donor's gender, ethnicity, drug and nicotine use history which would provide valuable leads to investigators.

The limitations in this study have been discussed, and these researchers have been careful to not overstate the findings or extrapolate the findings to casework. However, these researchers do feel strongly that based on the findings in this study there is



reason to be optimistic about future research in this area. A closer examination of specific bodily fluids using a larger sample size of donors where controls for variables like diet, use of cosmetics and nicotine are in place, is strongly recommended. If further research does show that the identification of key biologic and environmental markers and other identifiers can be determined, the forensic and investigative value of an unknown fingerprint will be significant. It will allow investigators to narrow their pool of suspects, apply additional forensic testing if warranted, and develop and analyze more discerning and relevant leads, which can lead to more expeditious resolution of cases that might otherwise languish for years.

# 5) References

Antoine, K. M., Mortazavi, S., Miller, A. D., & Miller, L. M. (2010). Chemical differences are observed in children's versus adults' latent fingerprints as a function of time. *Journal of Forensic Sciences*. 55(2), 513-518.

Archer, N. E., Charles, Y., Elliot, J. A., & Jickells, S. (2005). Changes in the Lipid Composition of Latent Fingerprint Residue with Time after Deposition on a Surface. *Forensic Science International*. 154(2-3), 224-39.

Asano, K. G., Bayne, C. K., Horsman, K. M., & Buchanan, M. V. (2002). Chemical composition of fingerprints for gender determination. *Journal of Forensic Sciences*. 47(4), 805-807.

Bright, N. J., Willson, T. R., Driscoll, D. J., Reddy, S. M., Webb, R. P., Bleay, S., ... & Bailey, M. J. (2013). Chemical changes exhibited by latent fingerprints after exposure to vacuum conditions. *Forensic Science International*. 230(1), 81-86.

Buchanan, M. V., Asano, K., & Bohanon, A. (1997). Chemical characterization of fingerprints from adults and children. *Forensic Evidence Analysis and Crime Scene Investigation*, 2941, 89-95.

Croxton, R.S., Baron M.G., Butler, D., Kent T. & Sears, V.G. (2006). Development of a GC-MS method for the simultaneous analysis of latent fingerprint components. *Journal of Forensic Sciences*. 51(6): p. 1329-1333.

Croxton, R. S., Baron, M. G., Butler, D., Kent, T., & Sears, V. G. (2010). Variation in amino acid and lipid composition of latent fingerprints. *Forensic Science International*, 199(1–3), 93–102. http://doi.org/10.1016/j.forsciint.2010.03.019

Frick, A.A., Chidlow, G., Lewis, S.W., van Bronswijk, W. (2015). Investigations into the initial composition of latent fingermark lipids by gas chromatography–mass spectrometry. *Forensic Science International*. 254(1): p. 133-147

Fullbright, S.I. (2014) Investigating Fingermark Aging by GC/MS: An Analysis of Current Methods. 1-37. George Mason University.



Girod, A., Ramotowski, R., & Weyermann, C. (2012). Composition of fingermark residue: A qualitative and quantitative review. *Forensic Science International*, 223(1–3), 10–24. http://doi.org/10.1016/j.forsciint.2012.05.018

Greene, R. S., Downing, D. T., Pochi, P. E., & Strauss, J. S. (1970). Anatomical variation in the amount and composition of human skin surface lipid. *Journal of Investigative Dermatology*. 54(3), 240-247.

Hartzell-Baguley, B., Hipp, R. E., Morgan, N. R., & Morgan, S. L. (2007). Chemical Composition of Latent Fingerprints by Gas Chromatography–Mass Spectrometry. An Experiment for an Instrumental Analysis Course. *Journal of Chemical Education*, 84(4), 689. http://doi.org/10.1021/ed084p689

Koenig, A., Girod, A., & Weyermann, C. (2011). Identification of Wax Esters in Latent Print Residues by Gas Chromatography-Mass Spectrometry and Their Potential Use as Aging Parameters. *Journal of Forensic Identification*, *61*(6), 652–676.

Kokatnur, M. G., Oalmann, M. C., Johnson, W. D., Malcom, G. T., & Strong, J. P. (1979). Fatty acid composition of human adipose tissue from two anatomical sites in a biracial community. The American Journal of Clinical Nutrition, 32(11), 2198–2205.

Ky, K. (2013). Method Validation of Extracting Fatty Acids from Latent Fingerprints by Thermal Desorption Gas Chromatography-Mass Spectrometry to Determine Personal Characteristics. 1-67. George Mason University.

Michalski, S., Shaler, R. & Dorman, F.L. (2013). The evaluation of fatty acid ratios in latent fingermarks by gas chromatography/mass spectrometry (GC/MS) analysis. Journal of Forensic Sciences. 58(S1): p. S215–S220.

Pappas, A., Fantasia, J., & Chen, T. (2013). Age and ethnic variations in sebaceous lipids. *Dermato-endocrinology*. 5(2), 319-324.

Pinto, D. (2014). "Qualitative Characterization of Fingermark Fatty Acid Residue by Liquid Extraction and Thermal Desorption Gas Chromatography Mass Spectrometry (TDUGC/MS)". 1-32. George Mason University.

Quintero Moreno, X. (2016). A comprehensive research study for Lipids found on Latent fingerprints and influence factors that affect their degradation mechanisms. 1-53. George Mason University.

Ramasastry, P., Downing, D. T., Pochi, P. E., & Strauss, J. S. (1970). Chemical composition of human skin surface lipids from birth to puberty. *Journal of Investigative Dermatology*. 54(2), 139-144.

Roane, K. R. (2005). The CSI Effect. U.S. News & World Report, 138(15), 48-54.

Shetage, S. S., Traynor, M. J., Brown, M. B., Raji, M., Graham-Kalio, D., & Chilcott, R. P. (2014). Effect of ethnicity, gender and age on the amount and composition of residual skin surface components derived from sebum, sweat and epidermal lipids. *Skin Research and Technology*, 20(1), 97–107. http://doi.org/10.1111/srt.12091



Wiseman, J. (2014). "The Application of Solid Extraction by Thermal Desorption GC/MS in the Determination of Physical Human Characteristics." 1-37. George Mason University.



# 6) Appendix

**Appendix A: Sub-contractor Technical Report** 

# **Final Report for SETCAF**

## Submitted to

George Mason University/ Department of Forensic Science Contract # E2027491

By



Contracted by:

# **George Mason University**

# **Technical POC:**

Dr. Michael Bowers 6315 Hillside Ct Suite F Columbia, MD 21046 michael.j.bowers@baesystems.com 410-312-5132

# Administrative POC:

Michael Frankhuizen 4301 North Fairfax Dr. Suite 800 Arlington, VA 22203 703-284-8437



# GMU Program Manager: Daniel O. Pinto, MSc.

The views and conclusions contained in this document are those of the authors.

Distribution Statement: The information contained in this document is considered BAE Systems Proprietary.

# **Executive Summary**

Sensitive site Exploitation through Trace Chemical Analysis of latent Fingerprints (SETCAF) is a technique for extracting chemical information from latent finger prints. Studies have shown that many attributes may be ascertained from the composition of fingerprint residues. This effort serves to provide a larger data pool than has been previously examined and to establish the methods for analyzing the samples. BAE Systems, as a subcontractor to George Mason University (GMU), has developed and refined a novel combination of instruments to perform Super-Critical Carbon Dioxide (SCCO2) extraction of latent fingerprints and subsequent analysis. We have shown that the instrument has adequate sensitivity and extraction/transfer efficiency to perform chemical analysis on latent finger prints. As stated in the Appendix A, we have performed this analysis on approximately 160 samples to date with minimal instrument down time owing to the robustness of the system. Further, we have demonstrated what we believe is the first non-destructive chemical analysis of a latent fingerprint through use of our novel fingerprint powder.



# **Technical Summary Program Overview**

<u>S</u>ensitive site <u>Exploitation through <u>T</u>race <u>C</u>hemical <u>A</u>nalysis of latent <u>F</u>ingerprints (SETCAF) is a technique for extracting chemical information from latent finger prints. Studies have shown that many attributes may be ascertained from the composition of fingerprint residues. This effort serves to provide a larger data pool than has been previously examined and to establish the methods for analyzing the samples. Two analysis instrumental techniques have been explored. Direct thermal desorption which has been deemed a hard extraction method and super critical carbon dioxide (SCCO2) which has been deemed a soft extraction technique. The difference between hard and soft techniques in this context is the destruction of the sample during analysis. Further</u>

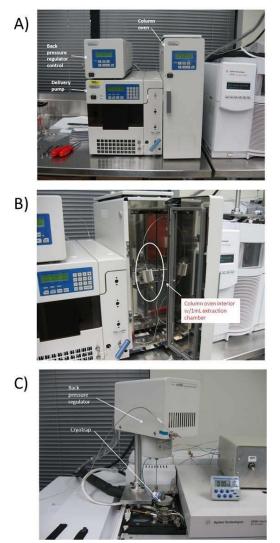
we have shown proof of principle that the SCCO2 extraction method may be used to lift latent finger prints visualized by BAE Systems proprietary fingerprint powder and perform a non-destructive chemical analysis of the prints. BAE Systems served as a subcontractor to George Mason University (GMU) on this effort and as such the following report summarizes the portion of the effort performed by BAE Systems.

# **Methods and Instrumentation Development**

# **SCCO2** Instrument

Over the course of the program, BAE Systems built and refined a custom system for performing the SCCO2 extraction and analysis of the finger prints via gas chromatography/mass spectrometry (GC/MS). Figure 2.2.1 shows the hybrid system which consists of a SCCO2 system from JASCO coupled to our Agilent GC/MS system via a cooled inlet system (Gerstel CIS4). As the SCCO2 leaves the backpressure regulator it goes from supercritical fluid to gas rapidly expanding its volume. Based on successfully modifying the CIS4 inlet for previous programs requiring a much larger volume of gas to pass through the inlet than the GC can manage, we applied the same principles and this allowed the use of the CIS4 to trap the target compounds for analysis via the GC/MS instrument Refinement of the instrument included automation of the process via

custom LabView script to increase both and reliability.



**Figure 2.2.1** SCCO2 system coupled to GC/MS via CIS4. A) is the JASCO reproducibility

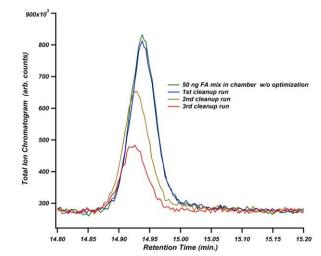


Early in the program, the focus shifted from small molecules to fatty acid analysis which proved to be challenging given our instrumentation suite. A change from a standard analysis column to a Stabilwax <sup>TM</sup> DA column from Restek allowed us to perform the fatty acid analysis without first functionalizing the fatty acid compounds as is typical standard analysis practice.

# **Instrument Parameters**

There are several instrument parameters inherent to a system like this that result from the coupling of the techniques. The inherent parameters for the extraction apparatus include extraction time, pressure, SCCO2 flow rate, extraction temperature and addition of a cosolvent. For the CIS4/GC/MS instrument, there are standard GC parameters such as oven temperature, flow rate, etc. that would affect the separation as well as cooled inlet parameters such as the inlet temperature and type of liner packing being used. The critical parameters will be discussed below.

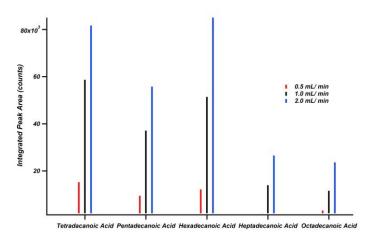
critical parameters will be discussed below. paramount importance for two key reasons. First, the sample sizes and available materials are small (micrograms). Even with the sensitivity of the GC/MS instrument finding and identifying small constituents of the complex mixture of compounds that make up finger print residues is a daunting task. Effectively extracting the compounds is critical. Second, inefficiency of transfer causes carryover problems which can result in lengthy clean up cycles to ensure that cross contamination of the samples is not skewing the instrumental results. **Figure 2.2.2.1** is a chromatogram, from early in the program that illustrates this problem



well. After introducing 50 ng of a fatty acid mixture into the extraction chamber,

subsequent blanks were run and overlaid. The first blank shows the same intensity as the initial exposed run and it slowly decays with subsequent runs. This presents a real problem for two reasons. It could take several hours of blanking to achieve a clean baseline and the potential sensitivity of the measurement is drastically reduced because clearly less than half of the sample was transferring from the extraction chamber to the GC/MS on the initial run.

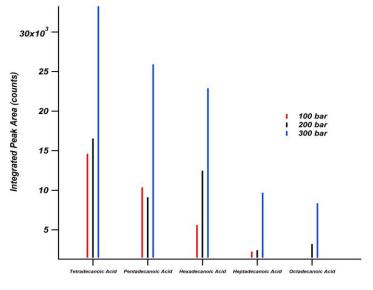
The parameters mentioned above





were all optimized to some degree but the largest improvements were found in extraction flow rate and pressure. **Figure 2.2.2.2** shows the initial recovery of several fatty acids from a 50 ng exposure with

increasing flow rate. The recovery improved by nearly a factor of 10 going from 0.5 ml/min to 2 ml/min. **Figure 2.2.2.3** summarizes the same experiment with increasing extraction pressure. Increasing the pressure made a dramatic improvement in recovery of the fatty acid analytes. This is likely due to the change in polarity that occurs in SCCO2 due to pressure. Smaller gains were made by increasing the extraction chamber oven temperature to 50 °C and increasing the extraction time. Addition of a co-solvent to the



SCCO2 also improved recovery. Addition of as little as  $100 \ \mu$ L of hexanes or isopropyl alcohol reduced carry-over percentages to single digits without the addition of the other gains described here. In the final method, direct cosolvent addition was omitted for reasons discussed later in **Section 2.2.3**. These experiments led to the final method parameters used to analyze the samples which are summarized below in **Table 2.2.2.1**.

The CIS4/GC/MS parameters were determined via the same type of methodology. The StabilwaxTM column provided adequate separation at near isothermal conditions for the fatty acid compounds so no complicated oven program was devised. To minimize column wear and bleed, a base starting temperature of 100 °C was implemented followed by a quick ramp at start to 240 °C at 30 °C degrees/minute. The key parameters for the analysis instrument fall with CIS4 inlet. The CIS4 is a cooled inlet that utilizes a packed (or open, depending on the

sample type) liner to collect **Table 2.2.2.1** Final instrument parameters used for sample samples. After surveying <sup>analysis.</sup>

different sorbent packed liners, it was determined that deactivated glass beads were the best candidate for a liner packing material. They provide surface area while not binding the fatty

Parameter	Value
Extraction time	14 min
Extraction pressure	250 bar
Extraction temperature	50 °C
Extraction flow rate	3 mL/min
CIS4 temperature	-50 °C init. ramp to 400 °C and hold
profile	11 min
CIS4 liner	Deactivated glass beads

acids strongly which leads to carry-over and peak smearing during desorption. Inlet temperature was kept to -50 °C. This proved sufficient to trap the fatty acid compounds of interest while minimizing the trapping of  $CO_2$  which can freeze in the liner clogging it and causing an over pressure condition. To accommodate the large volume of gas flowing through the liner, the GC pneumatic controls are bypassed during the sample



loading phase of the process. At the beginning of the analysis, CIS4 is ramped to 400 °C at 12 °C/sec to provide the sharpest injection possible. This is especially critical for compounds that have traditionally broad GC peaks.

## **Sample Preparation**

Fingerprint samples were provided by GMU on standard 1X3" glass slides. Samples were labeled with an identifying number but no information about the donor was present. Samples were stored in closed containers at ~4 °c. until analyzed. Originally, the prints were to be dusted with BAE Systems finger print powder and lifted for analysis; however, numerous attempts to find a tape that did not obscure the fatty acid signature or overwhelm the detector were unsuccessful. It was decided that the prints would be wiped from the slides using a moistened section of filter paper. The filter paper (Whitman grade QMA) was cut into small strips and folded in half to provide a sturdy wiping surface. The wipe was moistened with methanol to facilitate removing the oils from the glass. This methanol also serves as the co-solvent which provides cleaner extraction and minimizes carry-over in the system. It was determined that in most cases, a full fingerprint was too much sample for our system as the signal was saturated in the GC/MS and it would take several blank runs to clear down to baseline. Thus, a grid pattern was developed to

place the slide on where approximately 1 cm<sup>2</sup> of sample

was measured to extract from the glass (Figure 2.2.3.1).

At the operator's digression more or less sample area was collected depending on the apparent volume of residue in the fingerprint. The paper was then loaded into the extraction chamber for analysis.

Several precautions were taken to minimize the risk of sample contamination. This is of extreme importance due to the ubiquity of the analytes found in the natural environment. All solvents and filter paper sheets were

checked prior to use for contamination. All tools used to handle the samples were washed thoroughly with soap and water followed by a rinse in methanol. These items were checked periodically through the day to ensure integrity.

## **Analysis Methods**

Samples and blanks were analyzed using the above described instrumental methods. **Figure 2.2.4.1** is a typical total ion chromatogram (TIC) of a fingerprint sample. Several compounds were identified in addition to the chosen suite of fatty acid compounds. Most of them were related to cosmetics use in this particular case. In **Figure 2.2.4.1**, there are two chromatograms (red and blue trace) which are replicate swabs from the same finger

**Figure 2.2.3.1** Grid pattern for sample swabbing.

1"x 3" x 1 cm crosshair





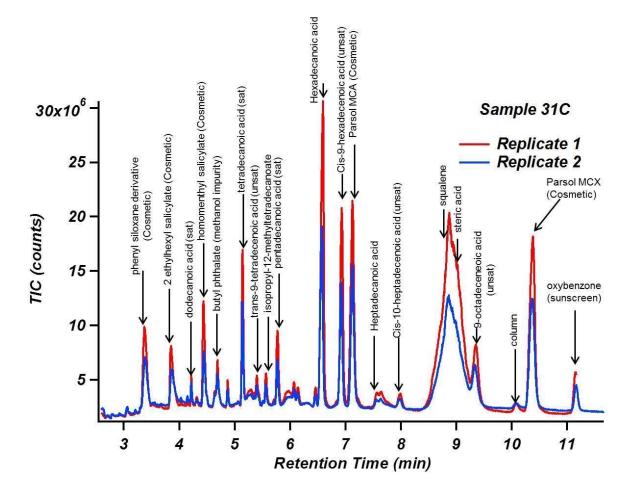


Figure 2.2.4.1 typical chromatogram of an extracted finger print. The two traces (red and blue) represent replicate swabs obtained from the same fingerprint.

print. The first 10 samples were run in duplicate to establish that there were no major differences (aside from sample loading) from one swab to the next. While the amplitudes of the peaks may vary dramatically, the relative ratio of the peaks remain similar. While ideally, a blank would be performed between each sample, time constraints forced blanking to occur at a minimum of after every third sample unless a sample showed excessively heavy loading. This was apparent to the operator as the samples were being analyzed and extra blanking was implemented as needed to ensure data integrity.

To minimize contributions of co-eluting species and column bleed, which was significant due to the nature of the column and the require operating temperature, single ion chromatograms were chosen as the source for peak integration. This is standard practice especially in cluttered chromatograms like that shown above in **Figure 2.2.4.1**. The list of compounds with respective retention times and integrated single ion are shown in **Table 2.2.4.1**. Integration of the peaks was performed primarily using manual integration within the Chem Station software suite. This was necessary due to the



irregular peak shapes that were prevalent in most of the samples. The irregular peak shapes and manual integrations led to issues with the calibration curves which will be discussed later.

In order to compare fingerprint samples, the instrument was calibrated via direct liquid injections of the fatty acid compounds. With the instrument response characterized, the transfer efficiency of the system was determined by replicating the sampling techniques used for introducing

the fingerprint samples. 5 μL of calibration
2.2.4.1 Compounds analyzed in this study using standard solution were placed on a methanol moistened filter paper swatch and \* analysis completed. The results are also summarized in Table 2.2.4.1. While the transfer efficiency is not critical to this analysis, it is provided so that future studies utilizing other instruments or improved techniques may be compared.

Using the calibration data, moles of each compound were calculated. Due to the extreme variation of the fingerprint samples with respect to size and loading, it was decided that rather than directly comparing the moles of each compound in

Compound Retention Ion Transfe Time (min) Efficie (amu) decanoic acid 3.5 129 100 % dodecanoic 4.2 200 65 % acid (lauric) 5.15 228 67 % tetradecanoic acid (myristic) pentadecanoic 5.8 242 56 % acid hexadecanoic 6.6 256 56 % acid (palmitic) palmitoleic 6.9 254 50 % acid 7.6 270 53 % heptadecanoic acid octadecanoic 8.9 284 53 % acid (steric) 9.4 18 % \* linoleic acid 264

the fingerprint samples a ratio of the fatty

acids with respect to the palmitic acid would serve as the value for comparison. Palmitic acid is a prominent peak that was easily integrated (reasonable peak shape) and was present in all samples. Using this ratio eliminates the sample size component of the equation as well as allows other researchers using different instruments to perform the same analysis and compare results directly.

# Samples Analyzed

# **Destructive Analyses**

Finger prints from approximately 150 individuals have been analyzed to date. In addition to these samples, 10 additional samples have been analyzed to provide storage life data. These samples are replicates of samples run earlier in the study to establish if storage time has a significant contribution to the results. Additionally, a set of 6 prints from the same individual were analyzed to determine if "smudges" are chemically equivalent to useable fingerprints. The raw peak integrations have been tabulated and provided to GMU for further analysis along with the necessary calibration data.

# Non-destructive Analysis Demonstration

To provide a proof of principle experiment and show that non-destructive analysis may be possible, a finger print was dusted with BAE Systems fingerprint powder, lifted with standard fingerprint lifting tape, and fixed to a piece of Whatman #1 filter paper. The

This document is a research report submitted to the U.S. Department of Justice. This report has not been published by the Department. Opinions or points of view expressed are those of the author(s) and do not necessarily reflect the official position or policies of the U.S. Department of Justice.

Table

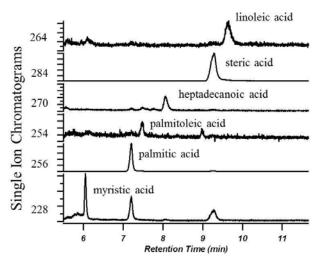


QMA filter paper was tried first but it was found that the tape did not stick very well to the paper resulting in the tape delaminating from the paper almost completely during



**Figure 2.3.2.1** Fingerprints dusted with BAE Systems fingerprint powder and extraction. **Figure 2.3.2.1** is an image of the dusted print affixed to the filter paper before and after being subjected to the SCCO2 extraction process. While tremendous amounts of adhesive were observed in some portions of the chromatogram, there were portions of the chromatogram that when examined in single ion mode (SIM) showed the expected

fatty acid peaks. **Figure 2.3.2.2** shows the SIM chromatograms with peaks labeled. To our knowledge, this is the first time any such analysis has been demonstrated. Future work here would revolve around finding the appropriate adhesive (possibly a thermosetting or UV curing material) that would allow a full analysis of not only the fatty acids but other chemical compounds of interest.



# Conclusions

BAE Systems has developed and refined a novel combination of instruments to perform SCCO2 extraction of latent fingerprints. We have shown that the instrument has adequate sensitivity and extraction/transfer efficiency to perform chemical analysis on



latent finger prints. We have performed this analysis on approximately 160 samples to date with minimal instrument down time owing to the robustness of the system. Further, we have demonstrated what we believe is the first non-destructive chemical analysis of a latent fingerprint through use of our novel fingerprint powder.