JOURNAL OF FORENSIC SCIENCES

J Forensic Sci, May 2011, Vol. 56, No. 3
doi: 10.1111/j.1556-4029.2011.01722.x
Available online at: onlinelibrary.wiley.com

TECHNICAL NOTE

CRIMINALISTICS

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Factors Affecting the Forensic Examination of Automotive Lubricating Oils*

ABSTRACT: Lubricating oil comparisons impact a variety of forensic investigations, including cases where oil was transferred from a suspect vehicle to the crime scene or victim. In this study, high-temperature gas chromatography/mass spectrometry was used to examine the influence of oil mixtures and oil changes over time on the comparison of known and questioned lubricating oils. Varying concentrations of oil mixtures were prepared and showed the potential for identifying individual components. Motor oils from 18 automobiles monitored over a 2-month period did not demonstrate significant changes in the chromatographic data. Chemometric analysis of motor oil mass spectral data provided little information regarding differentiation of, or changes in, the samples. Power steering fluid (PSF) from a naturally occurring leak collected from several locations was consistent with the PSF in the automobile's reservoir, and the PSF composition did not change over time.

KEYWORDS: forensic science, chemometrics, high-temperature gas chromatography/mass spectrometry, lubricating oils, motor oils, power steering fluid

The chemical analysis of oil is not a novel concept. The introduction of gas chromatography (GC) in the 1950s showcased an analytical technique that was highly similar to the distillation process utilized in the petroleum industry. As such, oil chemists and those involved in the petrochemical industry embraced this analytical development (1). The use of GC gained further foothold in the petroleum industry as the technique and detection methods improved. The development of the flame ionization detector, simulated distillation and high-temperature applications, the coupling of GC with the mass spectrometer (MS), and multidimensional GC techniques all resulted in GC becoming a primary analytical tool for the analysis of oils (1). The wide variety of GC techniques available has led to an abundance of petroleum product research including oil spill source identification, assessment of oil weathering, and the compositional analysis of oils (2–6). Research is not limited to the standard GC/MS approach. Specialized GC techniques, such as inverse GC and supersonic GC/MS, have both been used to examine the physical and chemical properties of oils (7,8). It should also be noted that other techniques, such as infrared spectroscopy (IR), X-ray fluorescence, atomic absorption spectrometry, and fluorescence spectroscopy, have been used to analyze oils (9–11). Much of the previous and current oil research is conducted by environmental agencies and/or the petroleum industry; however, the benefit of oil research extends beyond environmental forensics and the petroleum industry.

The detection and identification of various petroleum products, such as gasoline and lubricating oil, can have a significant impact on numerous forensic cases. Particularly, the comparison of lubricating oils has the potential to associate a crime scene sample to a suspect motor vehicle (12). Forensic scientists should remain aware of any potential factors that could impact comparisons so that reliable data is reported. Prior work by Zięba-Palus and Kościelniak (9) using a variety of techniques discusses the impact of motor oil additives and similar IR spectra on the ability to differentiate motor oils. Additional influences that should be considered, and that will serve as the focus of this research, include sample mixtures and changes in lubricating oils over time, both of which could affect the potential for sample differentiation.

The comparison of known and questioned samples is essential in a wide variety of forensic examinations. These comparisons can prove to be difficult for the examiner if either the known or the questioned sample has undergone significant changes in relation to one another. Past research has shown that unused lubricating oils can be distinguished from their used counterparts (13). Lubricating oils have the potential to undergo changes in composition, because of the wide array of operating conditions they are exposed to during their use in motor vehicles. The conversion of hydrocarbons into polycyclic aromatic hydrocarbons (PAHs) is one possible change that may occur in oil during use. PAHs that are more stable than their precursors can form when the hydrocarbon content of the oil is subjected to high temperatures, like those in an automobile engine (2,14). Owing to the increased stability of these compounds, they are monitored for oil fingerprinting and oil spill source identification (3,13). The formation of PAHs in motor oil tends to vary according to the amount of use the motor oil has undergone (e.g., the mileage), with unused motor oils showing little to no PAH presence; furthermore, different oil products can display different PAHs (2,10,15). Because of a variety of possible automobile operating conditions, the type of oil used, the degree to which the oil has been used, and the presence of PAHs or other changes in the

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*Presented in part at the 61st Annual Meeting of the American Academy of Forensic Sciences, February 15–21, 2009, in Denver, CO.

Received 15 Dec. 2009; and in revised form 26 April 2010; accepted 2 May 2010.

chemical composition of the oil may provide sufficient evidence to associate a lubricating oil sample with a specific automobile and determine that the sample is unique. Additionally, recognizing when, and if, changes in lubricating oils occur provides a timeline for analysts that conveys when a comparison of known and questioned samples is possible.

Mixtures of petroleum products may be discovered at crime scenes. The laboratory analysis of such mixtures can often be challenging, yielding results that are difficult to interpret. However, the identification of these mixtures is still possible through careful inspection of data. Distinguishing the components of mixtures may aid in the comparison of known and questioned samples and thus ensure that accurate conclusions are reached.

This paper will examine factors that can affect the analysis of lubricating oils. Using high-temperature gas chromatography/mass spectrometry (HTGC/MS), the effects of the presence of mixtures and sample variation over time on the comparison of known and questioned samples will be determined. The potential for sample differentiation, as a result of oil modification because of engine exposure, will also be explored. A naturally occurring lubricating oil leak will be used to simulate the type of sample that may be encountered at a crime scene and sent to the laboratory for analysis. Chromatographic data will be analyzed using qualitative chromatogram observations/comparisons, chromatogram smoothing techniques, and chemometrics. Multivariate statistics have been used extensively to analyze a wide variety of petroleum products ranging from diesel fuels to crude oils in an effort to identify key significant differences between highly similar samples and utilize these differences for sample differentiation/identification purposes (4,5,11,16–18). Owing to the use of these techniques in past research and based on the authors' past experiences, the authors felt that the analysis scheme would provide an effective means for achieving the research goals.

Methods

Exemplars

Two different unused motor oils, Valvoline All Climate 5W-30 (Valvoline; Ashland Inc., Lexington, KY), and Quaker State Synthetic Blend 10W-30 (Quaker State Corp., Irving, TX), were chosen as exemplars to depict the total ion chromatogram (TIC) of lubricating oil. Exemplar solutions were created in triplicate using 0.7% by weight oil in pentane (Burdick and Jackson, Muskegon, MI). The samples were subsequently analyzed on a PerkinElmer (PerkinElmer Inc., Wellesley, MA) Clarus 500 HTGC/MS.

Lubricating Oil Mixtures

A series of lubricating oils were mixed together to determine whether individual components of mixtures were detectable and to assess the affect of mixtures on making comparisons to exemplar samples. Three different mixtures of oils were used: (i) a mixture of two chromatographically similar conventional oils, Mobil 10W-40 Proven Performance (Mobil Oil Corporation, Fairfax, VA) and Pennzoil 5W-30 with Z-7 (Pennzoil Products Company, Houston, TX); (ii) a mixture of two chromatographically different conventional oils Kendall 10W-30 GT-1 High Performance (Kendall Motor Oil, Valley Forge, PA) and Castrol 20W-50 GTX Drive Hard (Castrol North America Incorporated, Wayne, NJ); and (iii) a mixture of a chromatographically different synthetic oil and conventional oil, Valvoline 10W-40 and Kendall 10W-30, respectively. Each of the mixtures was prepared, in triplicate, in ratios of 25:75, 50:50, and 75:25 by volume.

Automobile Samples

Motor oils from 18 different automobiles, shown in Table 1, were sampled three times over the course of June and July 2008, by swabbing the crankcase dipstick using a sterile cotton-tipped swab. Extra samples were taken as necessary, for example, if one of the automobiles received an oil change. The sticks of the cotton swabs were trimmed, and the swabs were placed into glass vials and capped.

Power steering fluid (PSF) from one automobile was also sampled in coordination with the motor oil during the months of June and July. The PSF was taken from the PSF reservoir using sterile cotton-tipped swabs. The PSF sampled in the reservoir was a mixture of unknown proportions of two different PSFs. Exemplar PSF samples were provided by the owner of the automobile for both fluids present in the mix.

TABLE 1—*Automobile sample number, make, model, year, oil information, and mileage information for each automobile used in the research sample set.*

Automobile Sample Base

Automobile Designation	Make	Model	Year	Oil Brand	Oil Type	Initial Mileage	Total Mileage Driven During Sample Period
1	Jeep	Grand Cherokee, 4x4, V8	2002	Mobil 1	5W-30 Synthetic	146,334	1616
2	Mercedes	S500	1998	Mobil 1	0W-40 Synthetic	168,592	4037
3	VW	Passat	2008	?	Synthetic	9611	2104
4	Toyota	Camry Hybrid	2007	?	5W-30 Synthetic	35,107	3893
5	Toyota	Prius	2005	Chevron	5W-30	41,343	1914
6	Subaru	Outback Legacy	1998	Exxon Superflo	10W-40	123,864	1897
7	Honda	CR-V	2003	Castrol	?	66,416	2218
8	Toyota	Corolla	2006	?	5W-30	18,711	1259
9	Ford	Focus	2004	?	?	77,217	1727
10	Ford	Focus	2002	Mobil	5W-30	106,348	2249
11	Ford	Mustang	2007	?	?	29,803	2313
12	Honda	Odyssey	2005	?	?	56,971	2363
13	Toyota	Sienna	2002	?	?	72,803	2157
14	BMW	528i	2000	?	?	54,732	1076
15	Ford	Crown Victoria	2008	Motorcraft	5W-20	8281	3480
16	Toyota	Matrix	2004	?	?	41,196	613
17	BMW	325xi	2004	Castrol	Synthetic	62,371	1291
18	Honda	Accord	2005	?	5W-20	52,695	2049

Lubricating oils were extracted from the cotton swabs by adding *c.* 4 mL of pentane to each vial so the heads of the swabs were fully immersed in solvent. After soaking for several minutes, the solution was removed from the vials, filtered using a Whatman® AUTOVIAL® syringeless filter device (Whatman Inc., Clifton, NJ) containing a 0.45-µm polytetrafluoroethylene membrane, and then analyzed by HTGC/MS. Concentrations of the extracts were adjusted as necessary if the initial TIC data indicated that the sample was too concentrated or dilute. Samples that were too dilute were concentrated by solvent evaporation under a stream of nitrogen.

Lubricating Oil Leak Analysis

One automobile in the sample set experienced a PSF leak at the beginning of the summer, and a spot formed on the asphalt underneath the vehicle. Sterile cotton-tipped swabs were used to collect PSF from the spot on the asphalt by rubbing the dry swab over the spot with enough pressure to ensure collection. PSF was also collected from the undercarriage of the vehicle where the PSF was dripping before reaching the ground. The undercarriage samples were also collected by rubbing dry, sterile cotton-tipped swabs over the areas of interest. Once the samples had been collected, the swabs were trimmed, placed into glass vials, and capped. Extractions were performed in the same manner as previously mentioned for lubricating oil samples. The PSF exemplar fluids provided by the owner of the automobile were used as a basis for comparison.

HTGC/MS Analysis

Experimental parameters are presented in Table 2. The detailed analysis procedure can be referenced to Reardon et al. (12).

Data Analysis

Chromatographic data was initially analyzed with PerkinElmer's TurboMass software, version 5.0. The Savitzky–Golay smoothing method was used with a corresponding window size (scans) of ±99 and a smooth number of 4. The following ions were used to produce the PAH extracted ion profiles (EIPs): *m/z* 178, 202, 228, 252, 276, and 278 (12).

TABLE 2—*Gas chromatograph and mass spectrometer parameters.*

Gas Chromatograph	
Column type	Aluminum Clad HT5 (SGE Incorporated, Austin, TX) 5% phenyl equiv./ polycarborane-siloxane 25 m, 0.22 mm i.d., 0.1 µm film
Injection volume	2 µL
Split ratio	30:1
PTV injector program	(1) 50–480°C, held for 2 min (2) Reduced to 400°C, held for 3 min
Temperature program	(1) 100°C for 1 min (2) 15°C/min to 400°C, held for 5 min
Total run time	26 min
Transfer line temperature	300°C
Carrier gas and flow rate	Hydrogen: 1 mL/min
Mass Spectrometer	
Mass analyzer	Quadrupole
Ionization mode	Electron impact
MS scan time	3–21 min
m/z scan range	50–550 *m/z*
Source temperature	300°C

PTV, programmable temperature vaporizer.

To prepare the data for chemometric analysis, the data were analyzed using Agilent's Chemstation (Agilent Technologies Inc, Santa Clara, CA) software. An algorithm was written that integrated the mass spectral data across the entire TIC for each sample. The abundance for each *m/z*, 50–550, was output in the form of a comma-separated value data file that was used for statistical analysis.

Statistics

Addinsoft's XLSTAT PRO® add-on (Addinsoft, Inc., New York, NY) for Microsoft Excel® (Microsoft Corporation, Redmond, WA) was used for multivariate statistical analysis of the data sets. To prepare the data sets for primary statistical analysis, each data set was normalized, which helped to remove absolute sample-to-sample variability (19). Normalization of each data set was accomplished by dividing each variable (i.e., abundance at each *m/z*) by a constant. The 2-norm constant was used and can be calculated as:

$$\text{2-norm} = \sqrt{\sum_{\text{j}=1}^{\text{nvars}} x_j^2}$$

After the data had been normalized, primary statistical analysis was completed in the form of agglomerative hierarchical clustering (HCA), principal component analysis (PCA), and discriminate analysis (DA).

Agglomerative Hierarchical Cluster Analysis

For this research, Euclidean distance measurements were used in coordination with Ward's linkage method. The responses for each *m/z* (i.e., 50–550) were used as variables and input into the XLSTAT® data analysis program. Dendrograms were generated by the software according to the dissimilarity between clusters.

Principal Component Analysis

XLSTAT® was used to perform PCA using the previously generated mass spectral data sets for each sample. By design, PCA results in a set of underlying factors where a large amount of variation is compressed into the early factors. However, beyond these first few factors, the remaining variation can be considered to be largely noise. Several tests are available to determine how many factors to retain, and in this case, a plot of the eigenvalues for the factors (Scree plot) indicated that the first four principal components were the most meaningful. The projection of the observations (observation scores) into this new four-dimensional principal component space were then used as inputs for subsequent statistical methods.

Discriminate Analysis

DA was performed using the sample scores from PCA as inputs. DA plots and confusion matrices were generated and the confusion matrix for cross-validation was considered in determining the accuracy of the data modeling. Predefined sample groups were established using two different criteria. The first DA plots were created for the entire data set using groups based upon the automobile from which the oil samples were taken during specific time frames. This set of plots was used to determine whether oil from different automobiles could be distinguished from other automobiles over the same time period. A second group of DA plots was generated for each individual automobile with predefined groups established according to the dates oil samples were taken from the automobiles. The second group of plots was designed to ascertain whether motor oil from the same automobile, sampled on different days, could be differentiated. Additionally, 95% confidence level ellipses were used to highlight sample groupings.

Results and Discussion

Motor Oil Exemplars

Chromatographic data obtained for the two exemplar oils (e.g., Valvoline 5W-30 All Climate and Quaker State 10W-30 Synthetic Blend) was used to create a basis of comparison for the oils gathered with the collection substrates. The chromatograms were subjected to the Savitzky–Golay smoothing algorithm, which removed peaks in the TIC to easily discern the shape of the unresolved envelope. The TICs for the two exemplars were easily distinguished by visual comparison.

While the two exemplars could be differentiated through a visual comparison of the unresolved envelope shape and the resolved hydrocarbons, replicate measurements of the exemplars were not 100% correlated. The smoothed and unsmoothed exemplar replicate chromatograms showed either subtle differences in the width of the unresolved envelope or in the shape and/or location of the apex (Figs 1 and 2). A plausible explanation for this disparity can be attributed to small variations in the MS scan rate from run to run. The chromatographic shape is slightly altered because of the MS scan "hitting" different points on the chromatogram. When the smoothing algorithm is employed, these changes become readily apparent.

These slight differences did not significantly detract from the overall chromatographic pattern of the envelope, yet exact "overlays" of the sample replicates were not possible. The same smoothing technique was employed by Reardon et al. (12) to differentiate between oils where minimal variation existed in the width of the unresolved envelope. However, the criteria for stating that two lubricating oils are distinguishable based upon the shape of the smoothed unresolved envelope must be re-examined. Owing to the differences seen among the exemplar replicates, the criteria previously established by Reardon et al. for sample differentiation can no longer be considered reliable. If samples are being compared using the smoothing algorithm and only subtle differences are seen between smoothed chromatograms, an analyst should only make the determination that the samples are consistent in regard to their chromatographic pattern. The analyst should not make the determination that the samples are of the same origin. Many different lubricating oils have highly similar chromatographic patterns (12). Therefore, discrimination based on subtle differences is not prudent.

Motor Oil Mixtures

HTGC/MS data generated from the analysis of motor oil mixtures conveyed the difficulties inherent in the examination of

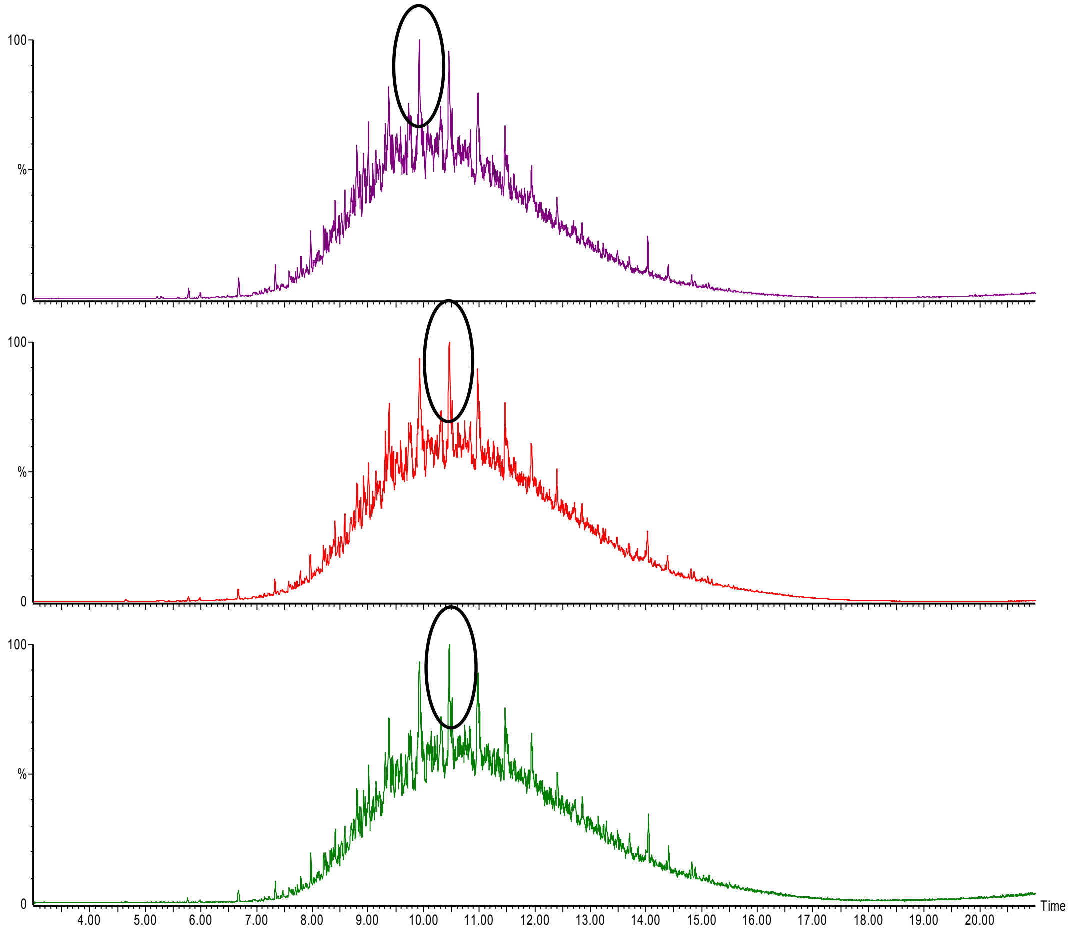

FIG. 1—*Valvoline 5W-30 exemplar replicates showing a shift in the maximum apex peak.*

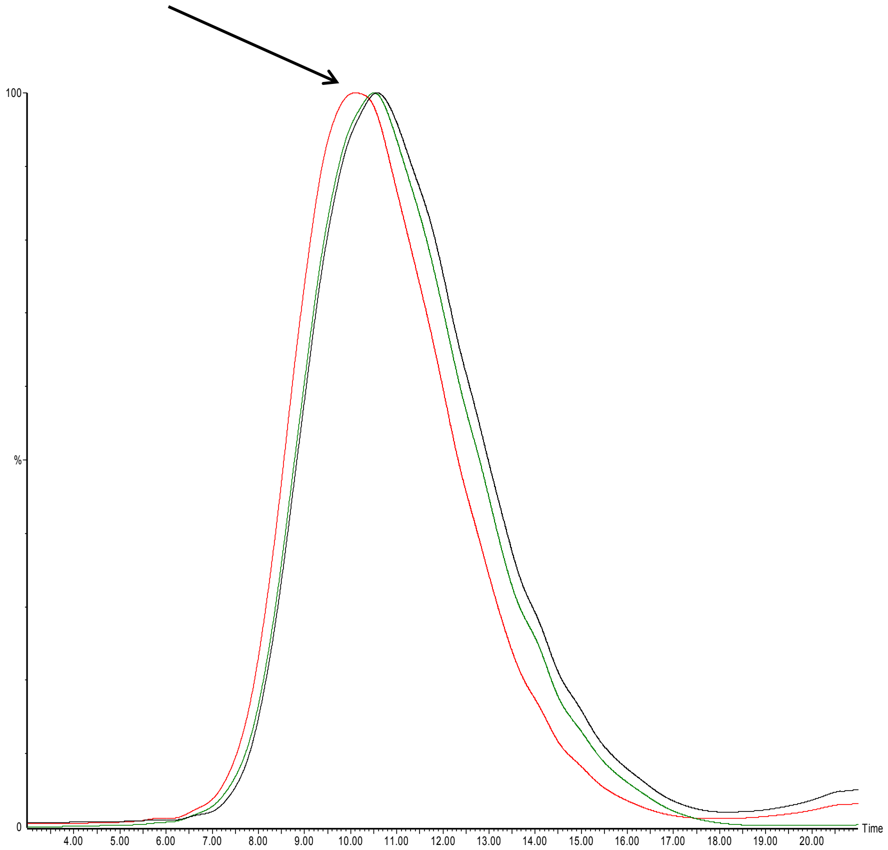

FIG. 2—*Overlay of smoothed Valvoline 5W-30 chromatograms seen in Fig. 1. Arrow denotes the chromatogram with a different maximum apex peak.*

lubricating oil mixtures, as well as the possibility of identifying a sample as a mixed-oil sample. These difficulties should be considered in the assessment of sample composition and during a questioned and known sample comparison.

Data for each of the three mixtures were analyzed, but chromatographic data for the mixture of two similar conventional motor oils were the most difficult to interpret. The two oils used to make the mixture differed in the width of the unresolved envelope and in the presence of two spiking hydrocarbons (at *c.* 13.8 min) that were in the Mobil oil exemplar. While the two oils were similar, this variation between the samples allowed for their differentiation (Fig. 3). As demonstrated in the TICs of the mixtures (Fig. 4), variations in the width of the unresolved envelope were difficult to account for because of the consistency in the shape of the unresolved envelope. Yet, the two spiking hydrocarbons from the Mobil oil exemplar were present in varying intensities in each of the mixtures.

Chromatographic data for the mixture of two different conventional oils and the mixture of synthetic and conventional oils were easier to interpret than data for the two similar conventional oils. Both mixtures containing chromatographically different oils were easily distinguished through comparison of exemplar TICs. The lack of similarity between the exemplar oils in these mixtures allowed for easier recognition of the individual components. The 25:75 and 75:25 mixture predominantly resembled the TIC of the major constituent. Nonetheless, in some cases, identification of the minor component is possible. Detection of mixture constituents in the 50:50 mixes is also possible; yet, one oil may still be unequally represented in the chromatogram. An overlay of the smoothed TICs for the mixture containing synthetic oil and conventional oil can be seen in Fig. 5. The chromatogram overlay suggests that the 25:75 and 50:50 mixtures can be reliably distinguished from both of the exemplars. The 75:25 mixture cannot be differentiated from the Valvoline Durablend exemplar because the overall chromatographic shape is consistent. In this case, the determination that the samples were comprised of multiple oils could be made in two of the three mixture ratios.

Data from each of the three mixtures formed, in varying ratios, imply that the ability to make a determination that an oil sample is a mixture of multiple oils is highly situational and dependent on the constituents, and their relative amounts, in the mixture. Determining that a sample is a mixture requires that exemplar oils be submitted and that the exemplar oils be distinguishable. Even though the classification of a sample mixture is not possible in every situation, laboratory personnel should still consider the possibility of mixtures when a lubricating oil sample is submitted for analysis. If the sample appears to contain contributions from more than one exemplar sample, a mixture series of the suspected oils should be prepared in varying ratios for analysis.

Automobile Samples: Polycyclic Aromatic Hydrocarbons

Owing to previously published material (12,15), the authors originally believed that the formation of PAHs in the different motor oils would be directly linked with the amount of use the oil had experienced and that any increase in the amount of PAHs would occur within definable intervals. Based upon this original hypothesis, the authors also anticipated that variation in the amount and type of PAHs found in motor oil would provide a basis for identification of that oil. Unfortunately, examination of the PAH EIPs, for each sample taken from the automobiles did not support either

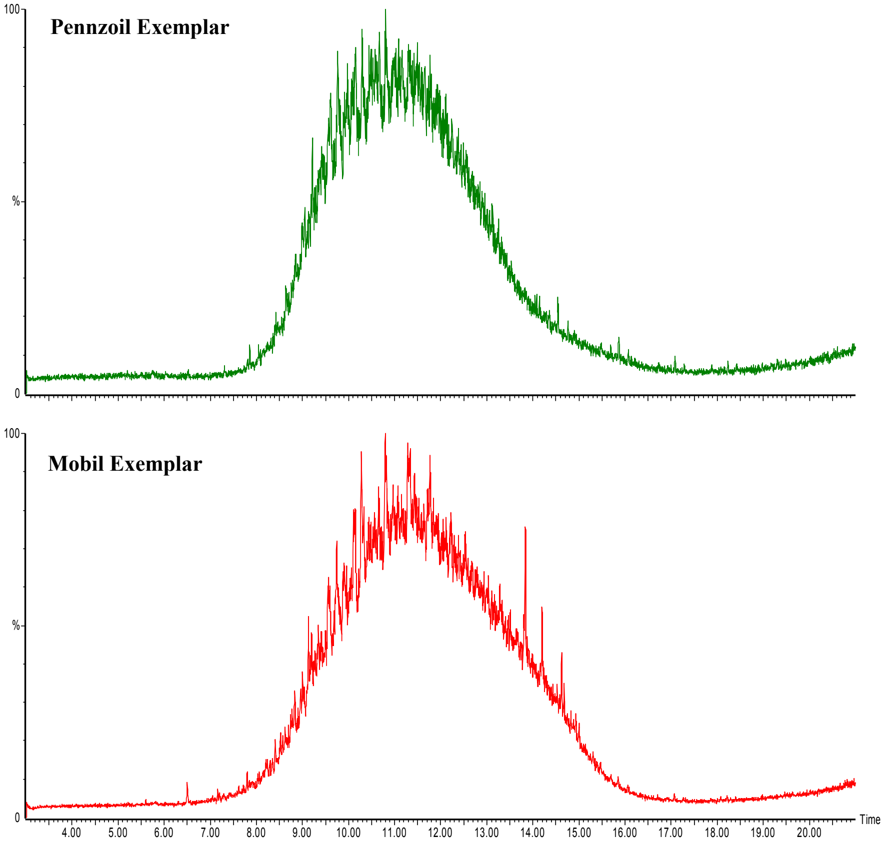

FIG. 3—*Total ion chromatograms for one mixture that used Mobil and Pennzoil oils.*

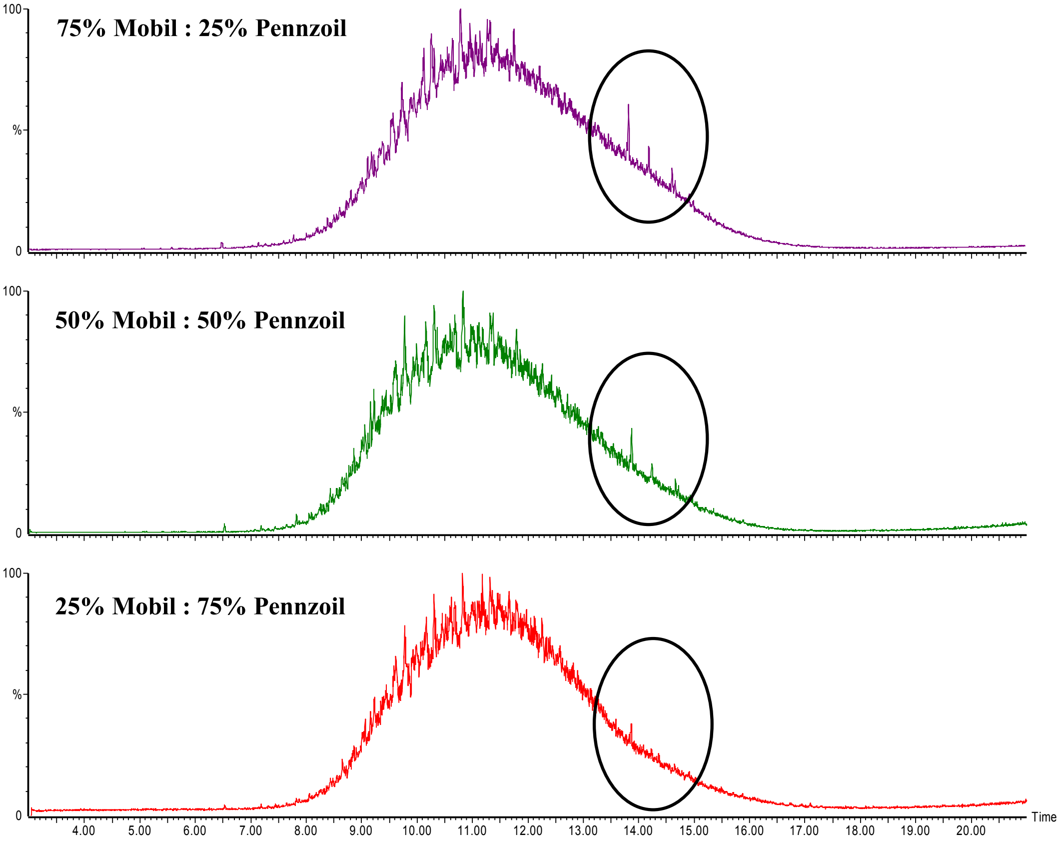

FIG. 4—*Total ion chromatograms showing varying intensities of spiking hydrocarbons from the Mobil exemplar in the Mobil–Pennzoil mixture.*

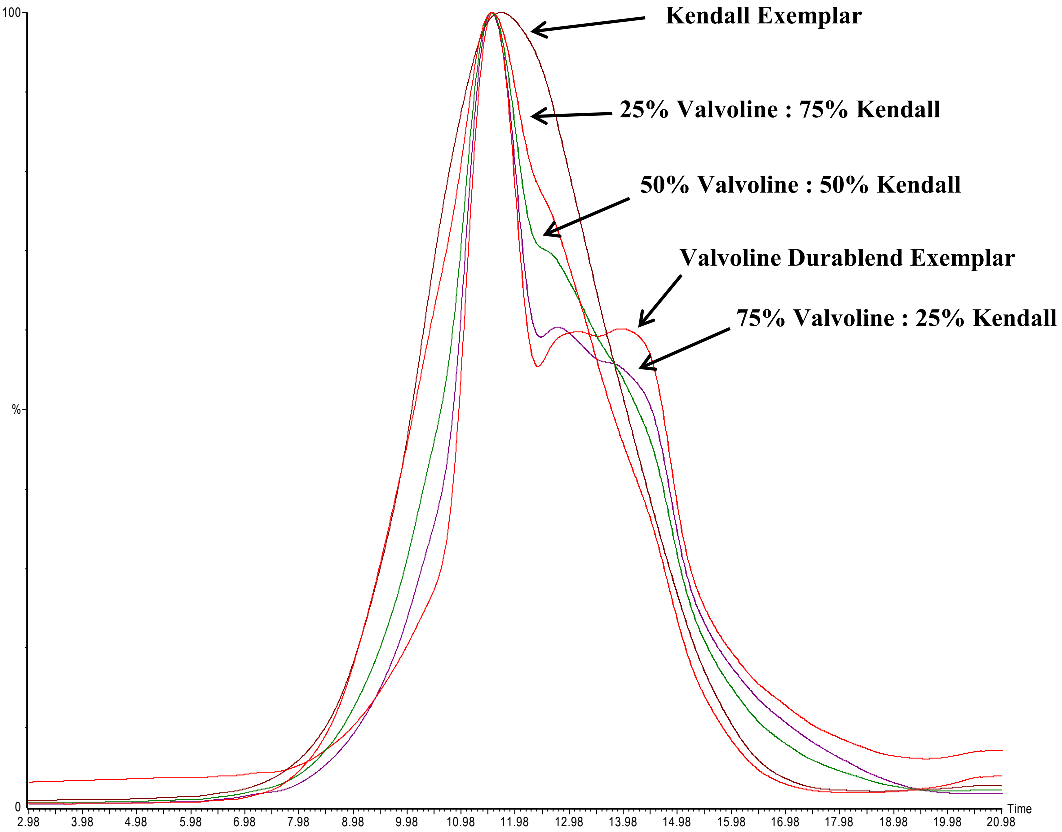

FIG. 5—*Overlay of smoothed total ion chromatograms for the mixture containing a synthetic oil and a conventional oil.*

hypothesis as being valid. PAHs were detected in at least one sample from each of the 18 automobiles, with anthracene, fluoranthene, and pyrene being the most prominent.

Figure 6 shows the PAH EIPs for a single automobile over the course of the summer. Formation of PAHs in the oils was not fully dependent on the degree of use (e.g., mileage), the type of oil, or the type of automobile. The only trend that was consistently displayed in the PAH EIPs was that the amount of PAHs detected in an automobile's motor oil was drastically reduced after the automobile experienced an oil change and that the PAHs detected in the EIPs remained consistent from sampling to sampling.

Variation did exist in the absolute amount of PAHs detected in the oils from different automobiles, within the same automobile for different sample dates, and within sample replicates. This observed variability was likely a reflection of concentration differences attributable to the sampling technique. The concentration of oil present in each extract was unknown, as the amount of oil retained by the cotton swabs used for collection was highly variable. Differences in concentration would lead to the differences seen in the PAH signal intensity within the sample replicates and may also explain variation present between automobiles and between different samples taken from the same automobile. The discrimination of motor oils from the analysis of PAH content was not possible using methodology described in this study.

Automobile Samples: Sampling Over Time and Differentiation

Although this study produced unanticipated results, the observed lack of change over time in the oil samples is beneficial to the forensic community. Chromatograms corresponding to automobile 6, which underwent an oil change during the sample period, are representative of the trends seen throughout the entire data set and can be seen in Fig. 7. A visual comparison of these TICs shows that there is little to no variation in the shape and width of the unresolved envelope or in the presence of spiking hydrocarbons along the unresolved envelope. The lack of variation in the oils over time allows for oil samples from a suspected source vehicle to be gathered weeks after a questioned sample was deposited at a crime scene, without concern that the oil from the vehicle underwent significant changes since fluid deposition at the scene. However, it is important to keep in mind that different motor oil samples with highly similar chromatographic patterns (i.e., relatively the same width and shape of unresolved envelopes and similar distribution of spiking hydrocarbons) cannot be distinguished through an analysis of the TIC.

Automobile Samples: Chemometrics

An attempt at differentiating the oils and at confirming the lack of changes in the oil over time was made by using chemometrics to analyze the integrated mass spectra of the oil samples. Mass spectral data were chosen because of the lack of resolved peaks in the chromatograms, which can cause difficulty when interpreting data based on retention times, and because of the lack of significant differences inherent in the chromatograms for a large proportion of the oils sampled. Additionally, analyzing lubricating oils according to their total mass spectra is not a common practice; consequently, a method exploiting this data may lead to the introduction of a new viable technique that can be applied in the forensic laboratory. The authors believed unique mass spectra may be produced for the oils from the different automobiles owing to differences in automobile operating conditions or formulaic differences in the oils themselves. Unfortunately, the potential for sample discrimination,

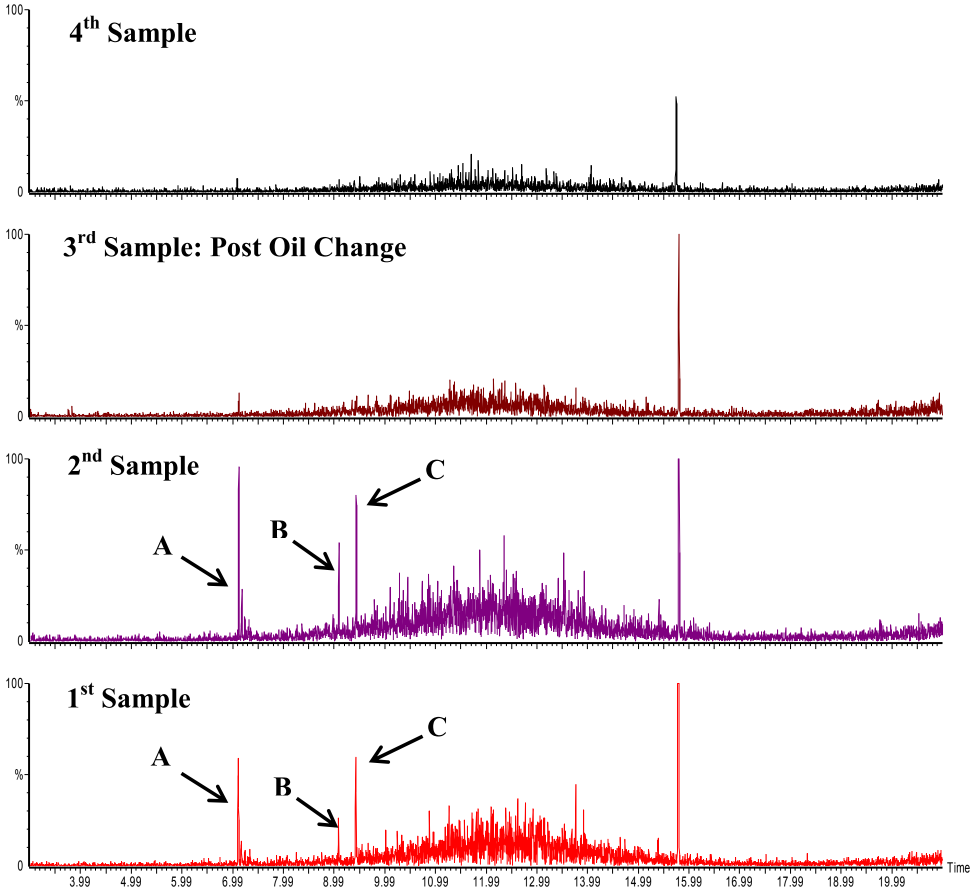

FIG. 6—*Automobile 6 polycyclic aromatic hydrocarbon extracted ion profiles. Labeled peaks are (A) anthracene, (B) fluoranthene, and (C) pyrene. Refer to Table 1 for the total mileage driven by automobile 6 during the study. The third sample was taken 125 miles post-oil change.*

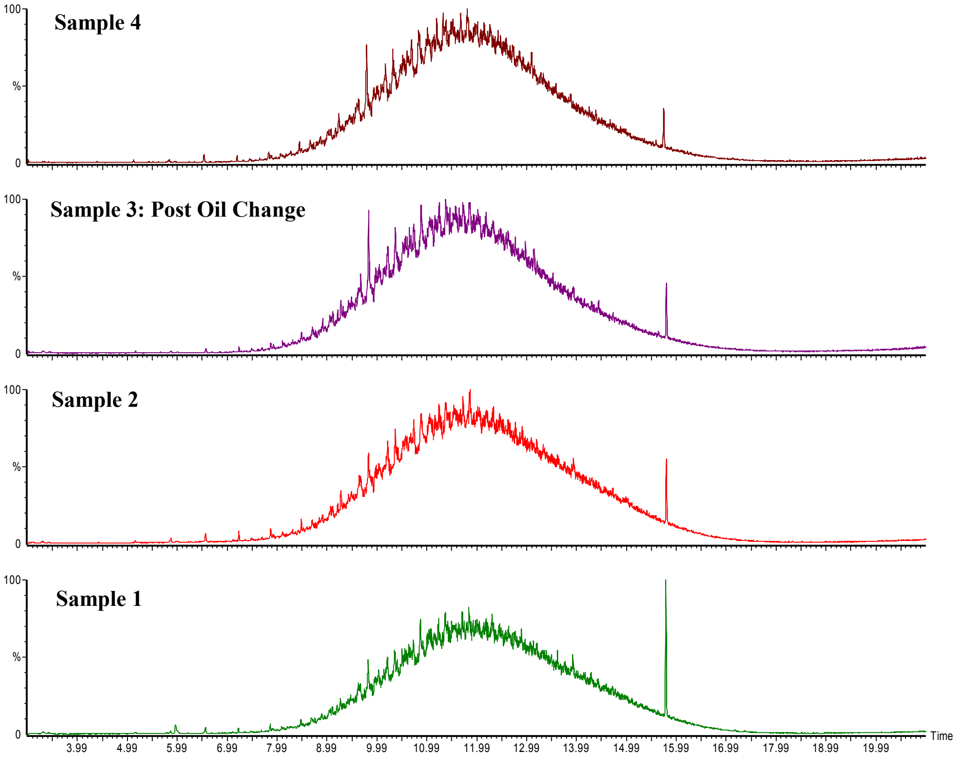

FIG. 7—*Automobile 6 total ion chromatograms for each oil sampling during the summer. Refer to Table 1 for the total mileage driven by automobile 6 during the study. The third sample was taken 125 miles post-oil change.*

whether by sample date or automobile, using HCA, PCA, and DA was largely unsuccessful. Each of the statistical algorithms used to analyze the data attempted to place data into distinct groups by focusing on the elements in the data set that most accurately allowed for differentiation of different samples and clustering of similar samples. Ideally, each of the 18 automobiles would be grouped separately, and the within-class distribution of the samples would be affected by the changes in the motor oils over time. Each statistical technique was unable to group similar samples (e.g., from the same automobile and replicates) reliably or differentiate between samples that were easily distinguished on visual examination of the TICs.

The HCA technique employed during the course of this research was determined to be ineffective and unsuitable for the generated data sets. Although the authors originally thought that analysis of the data with HCA would establish a gross representation of sample distribution within the data set, the dendrograms produced as a result of HCA failed to provide meaningful data. The dendrograms produced by the statistical algorithm were based upon the dissimilarity and similarity of the summed mass spectra. The algorithm did not apply any weight to individual variables (ions) that may be responsible for describing meaningful variation in the data set. Each of the samples' mass spectra was comprised of highly similar hydrocarbon fragments and ions associated with column bleed, both of which led to the "confusion" in the HCA algorithm. Because the algorithm was unable to filter out those variables that have little effect on sample differentiation, replicates from one automobile were often considered to be similar to a completely different automobile on a different day instead of being most similar to samples from the same automobile taken on the same day. As such, these results did not provide any meaningful insight into the data set.

TABLE 3—*Summary data for the first set of discriminate analysis plots highlighting cross-validation values and determination of unique samples.*

Sample Time Frame	Estimation Sample (%)	Cross-Validation (%)	Unique Automobiles
06/03/2008–06/05/2008	55.56	38.89	2,3
06/23/2008–06/24/2008	55.56	29.63	2
07/11/2008–07/15/2008	31.48	18.52	None

PCA is an effective technique for reducing the dimensionality of a data set, thereby helping to filter out those variables that do not have a significant impact on the variability within the data. The data from the generated score plots were used as input for DA. By using the PCA data in this manner, the linear discriminate function generated in DA should be based upon meaningful variation in the data set. It should be noted that the score plots were only used to generate DA input data and that they were not considered in their ability to show sample distribution.

DA was first used in an attempt to determine whether motor oil samples taken from different automobiles, during the same time frame, could be differentiated. Three sample periods were established to investigate this possibility. Each of the three sample periods encompassed all 18 automobiles and corresponded to the time frames where the first, second, third, and any immediate post-oil

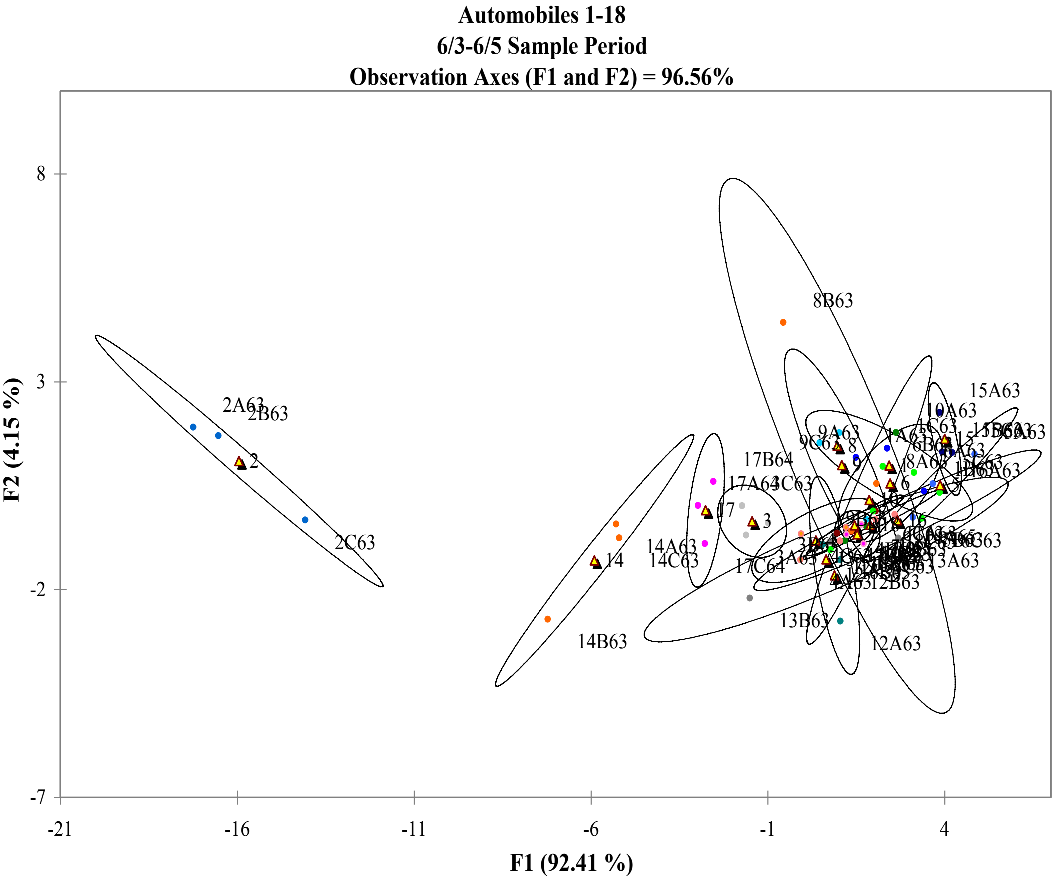

FIG. 8—*Discriminate analysis plot, with 95% confidence ellipses, for all 18 automobiles after the 6/3–6/5 sample period showing that Automobile 2 is "unique." Triangles denote sample centroids.*

change samples, between sampling dates, were taken. Unfortunately, the discrimination seen between automobiles on these plots was poor. This is demonstrated in Table 3 by the low cross-validation values that accompany the DA plots for each given sample period. The low cross-validation numbers illustrate that the discriminate function outlined by the DA statistical method did not allow for accurate sample clusters. Despite the low cross-validation values, an automobile was considered unique if all samples/replicates were classified correctly and if no other sample was misclassified to that particular automobile's group. Using these criteria, there were a very small number of automobiles that could be distinguished from all others during certain sample periods. Table 3 shows that automobiles 2 and 3 were both discriminated during the 6/3–6/5 sample period and that automobile 2 was the only unique automobile during the 6/23–6/24 sample period. No automobile was determined to be unique during the final sample period spanning from 7/11 to 7/15. The DA plot shown in Fig. 8 also shows that automobile 2 was distinct when compared to the 18 automobiles during the 6/3–6/5 sample period; however, automobile 3 did not appear as a distinct sample group even though the sample fit the criteria for a "unique automobile" in the confusion matrix. Automobile 2 had several qualitative factors that may have led to its individualization. Automobile 2 was the only Mercedes in the sample group, the only sample that was known to use 0W-40 oil, and the automobile that drove the most miles during the research. The only qualitative factor that may have led to automobile 3 being indentified during the first sample date was that this automobile was the only Volkswagen in the sample set. While qualitative factors for these two automobiles do exist, they did not differentiate these automobiles from the remaining automobiles during every sample period. Further research will have to be conducted to determine whether the individualization seen in the first DA plots was actually random or was based upon predictable attributes.

A second set of DA plots was generated to determine whether motor oil from the same automobile could be differentiated on different days, which would highlight any changes in the motor oil samples that were occurring over time. DA plots were generated for each of the 18 automobiles with groupings based upon the dates that motor oil samples were taken. The data from the second set of plots show that motor oil samples taken from the same automobile on different days was largely indistinguishable; however, a limited number of the automobiles had unique sample dates. Sample dates were considered unique according to the same criteria previously mentioned. Table 4 highlights the five automobiles that had unique sample dates as well as the cross-validation values for each sample plot. A DA plot for automobile 2, showing all sample dates, can be seen in Fig. 9. Again, although the cross-validation values were low for each of the plots, certain dates were classified correctly 100% of the time without any accompanying misclassifications. Three of the five automobiles that had unique sample dates had undergone an oil change during the course of the study. The unique dates that were noted for these three automobiles did not all coincide with the sample date that was closest to the date of the oil change, where changes in the PAH content of the samples occurred. Furthermore, all of the automobiles that underwent an oil change did not show a unique sample date, and some automobiles that did not undergo an oil change showed unique sample dates. This suggests that an oil change has a minor affect on sample discrimination. There was no readily definable pattern, or criteria, seen in the data that explained why certain sample dates for certain automobiles were determined to be unique. It should also be noted that even though certain sample dates were different according to DA, there were no qualitative differences seen in the sample TICs that would allow for differentiation. The final results from the second set of DA plots were similar to the first set of DA plots in that the unique sample dates for each of the automobiles may be an

TABLE 4—*Summary data for the second set of discriminate analysis plots showing post-oil change sample dates, cross-validation values, and unique sample dates.*

Automobile Number	Post-Oil Change Sample Date	Estimation Sample (%)	Cross-Validation (%)	Unique Date(s)
1	None	66.67	33.33	None
2	06/10/2008	83.33	58.33	07/14/2008
3	06/09/2008	75.00	41.67	06/09/2008
4	06/23/2009	77.78	55.56	None
5	None	88.89	44.44	None
6	07/01/2008	100.00	75.00	07/14/2008
7	06/23/2008	88.89	55.56	None
8	None	66.67	22.22	None
9	None	88.89	44.44	None
10	07/14/2009	77.78	55.56	None
11	None	100.00	55.56	None
12	None	88.89	66.67	None
13	None	77.78	11.11	None
14	None	100.00	88.89	06/03/2008
15	None	100.00	88.89	07/14/2008
16	None	77.78	55.56	None
17	None	66.67	33.33	None
18	06/23/2008	100.00	55.56	None

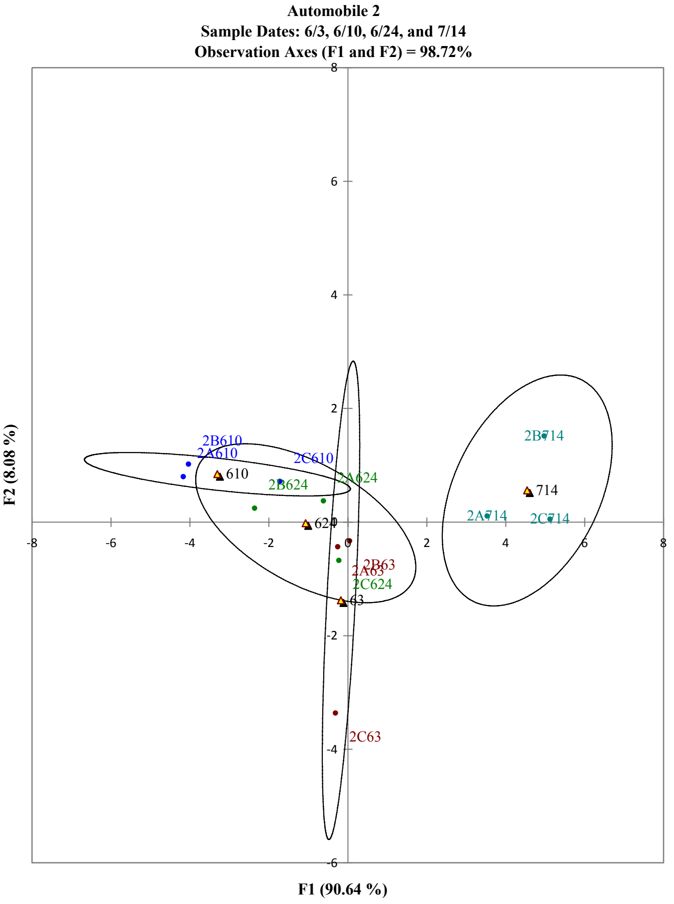

FIG. 9—*Discriminate analysis plot, with 95% confidence ellipses, for all sample dates for Automobile 2. Sample date 7/14 is "unique." Triangles denote sample centroids.*

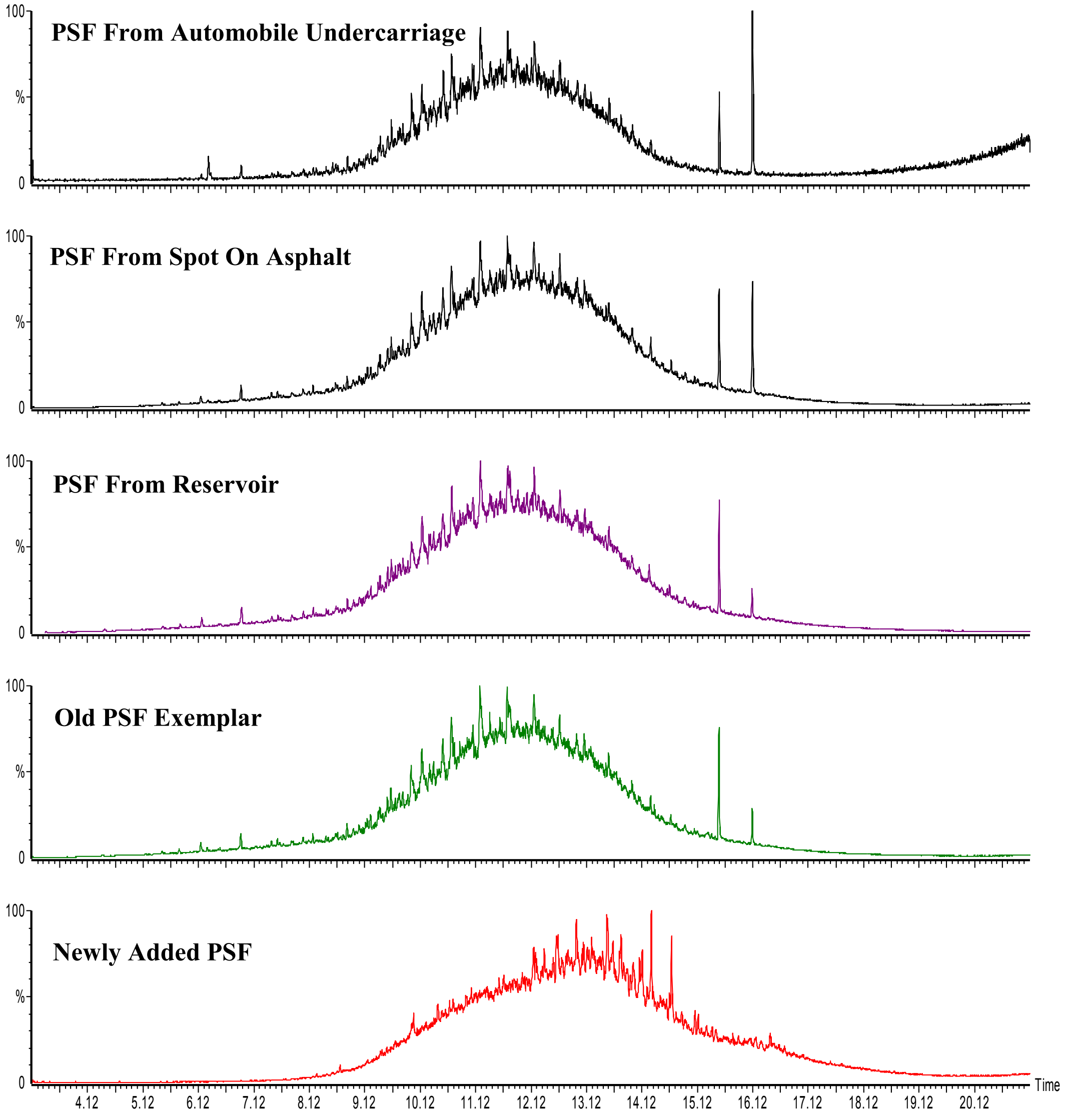

FIG. 10—*Total ion chromatograms for power steering fluid (PSF) samples taken from various locations. Note the consistency between all samples except the newly added PSF.*

unpredictable event that cannot be characterized. Further work should be carried out to ascertain whether or not sample discrimination is random or based on definable properties.

Power Steering Fluid

PSF samples gathered from an automobile leaking fluid simulated the type of evidence that may be found at a crime scene. Upon analysis of the chromatographic data for the PSF samples, several promising conclusions were made regarding collection of fluid leaks and the comparison of known and questioned samples. Examination of the TICs, for the exemplar PSF originally leaking from the reservoir and the new PSF that was added to the reservoir after the leak was discovered, showed that the two different PSF samples were easily distinguishable.

PSF samples gathered from a leak spot underneath the automobile and samples gathered from the undercarriage of the automobile were shown to be nearly identical to PSF samples gathered from the PSF reservoir of the automobile responsible for the leak. The TICs and smoothed TICs for a PSF spot sample and a PSF sample from the undercarriage of the automobile were compared to the old PSF, reservoir PSF, the new PSF added to the reservoir, and to each other (Figs 10 and 11). The consistency between all of the samples, with regard to the shape of the unresolved envelope and the spiking hydrocarbons seen in the TICs, indicated the automobile as a potential source of the fluid leak. The data also conveyed that the amount of new PSF fluid added to the reservoir was minimal, as all of the reservoir samples and leak samples were indistinguishable from the old PSF fluid.

PSF fluid was continually sampled from the reservoir of the automobile to determine whether more of the new PSF would "appear" in the chromatograms as the fluid was processed by the automobile and to determine whether any changes occurred in the fluid over time that would lead to inconsistencies between the reservoir samples and the leak samples. No noticeable changes were seen in the chromatograms during the sample period, and the influence of the new PSF remained undetected in every sample. The lack of changes seen in the chromatograms indicated that the chemical composition of the PSF was not influenced as the fluid traversed the power steering column of the automobile. Because of this, leak samples taken *c.* 5 weeks prior to the last reservoir sampling remained consistent with all reservoir PSF samples.

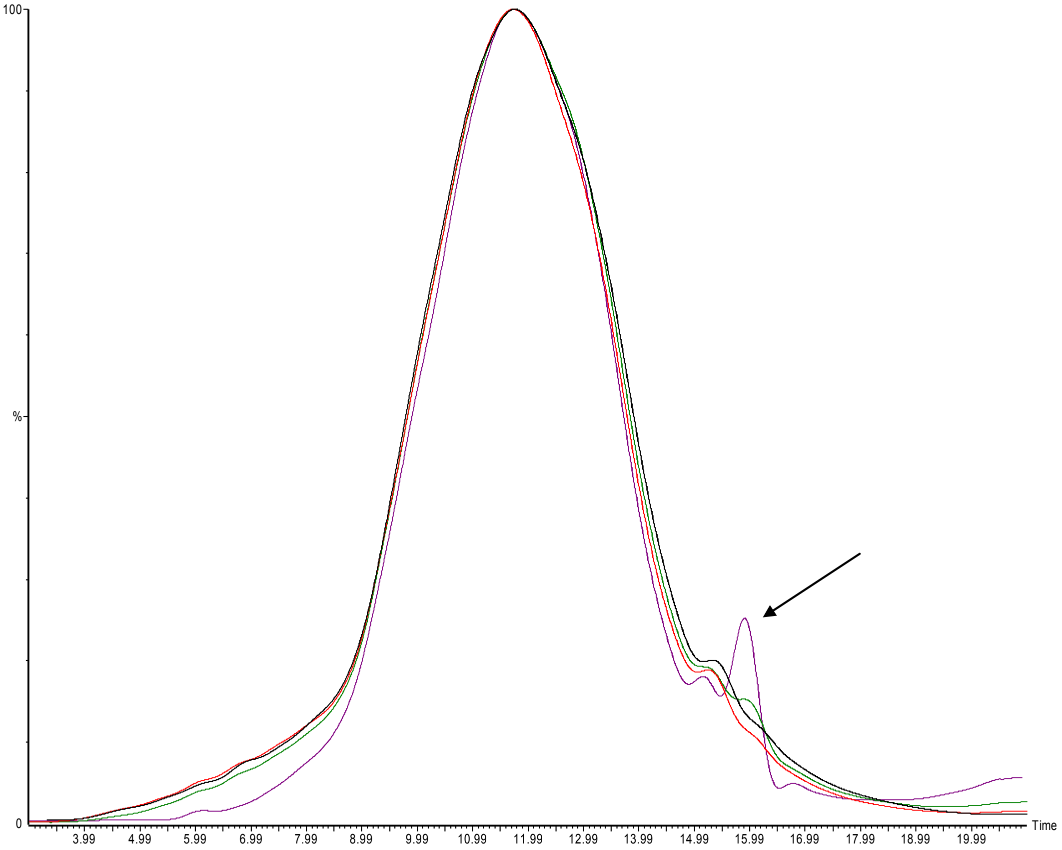

FIG. 11—*Smooth overlaid total ion chromatograms from Fig. 10, excluding the new power steering fluid, showing a high degree of correlation. Arrow denotes a filter peak.*

Data from the analysis of the PSF samples support the conclusion that lubricating oil samples collected from a crime scene can be correlated with exemplar samples from suspected source automobiles. Collection of the leak samples can occur from spots on the ground or from any portion of the automobile undercarriage that contains fluid from the leak location. When possible, crime scene investigators should collect samples from both locations to ensure consistency between the two, which increases the probability that the suspect automobile is the source of the fluid leak. While suspect exemplar samples should be collected in a timely manner, correlation within the PSF and motor oils over time conveys that successful comparisons could still be possible if samples cannot be gathered immediately.

Conclusions

HTGC/MS was used to analyze oil mixtures and monitor oils sampled over a 2-month period. The analysis of the oil mixtures provided valuable information concerning the identification of a sample as being a multicomponent mixture. The analysis of the oil mixtures demonstrated that identification of a sample as a mixture is possible, yet highly situational, as long as exemplars are provided for each of the oils in the mixture.

There was no variability in the PAHs identified in the automobile samples and no discernable trend in PAH formation. It was determined that PAHs cannot be used as a basis for the differentiation of motor oils with the methodology employed here.

Past research has suggested that unique operating conditions in automobiles will impart unique characteristics to motor oil and/or cause the oil to change over time. The TICs for each oil sample, taken at various increments, were consistent throughout the duration of the summer. Chemometric analysis of the integrated mass spectral data did not yield results that accurately described samples taken from one automobile over time, nor did the algorithms allow for differentiation of samples taken from different automobiles. The lack of change in the sample TICs over time and the inability of the statistical algorithms to describe the data sets reliably implies that (i) lubricating oil samples remain consistent in chemical nature over time, regardless of their environment, (ii) mass spectral data from the HTGC/MS cannot be used as a means to discriminate between lubricating oil samples, and (iii) lubricating oil samples, including those of different brands and taken from different automobiles, are highly similar in regard to their chemical composition. This high degree of similarity may be the factor that most accounts for the inability of the statistical methods to classify each of the samples accurately, regardless of the criteria used for clustering. It should be noted that the aforementioned conclusions are the result of the specific methodologies used during this research. Using different techniques, instrumentation, and/or methodologies may have the potential to show significant trends in lubricating oil data that were not discovered over the course of this research.

Analysis of the PSF fluid gathered from a natural automobile leak produced promising results for the forensic community. PSF samples gathered from a leak spot underneath the automobile, the PSF reservoir, and the undercarriage of the automobile displayed nearly identical unresolved envelope characteristics. Additionally, PSF sampled throughout the course of the summer remained highly correlated with all PSF samples taken at an earlier date. The obtained data offer support for the conclusion that lubricating oil samples can be associated with an exemplar vehicle and that the chemical composition of the fluid will remain consistent over extended time intervals.

To fully understand the potential for discriminating lubricating oils and associating a specific lubricating oil with a suspect vehicle, future HTGC/MS research should be pursued. Further studies that may be warranted include (i) the use of chemometrics in conjunction with retention time data from the TIC (18), (ii) the use of SIM mode to increase the sensitivity of PAH detection, (iii) lower electron impact voltages in the MS to create larger analyte fragments, (iv) analyzing more "mock case" scenarios, and (v) developing blind tests for evaluation. Research in the aforementioned areas will provide greater insight into the forensic analysis of lubricating oils and the conclusions that can be reached from their examination. Furthermore, the information garnered during the course of this, and future, research will have implications beyond the analysis of lubricating oils, as several of the techniques mentioned are currently being utilized to analyze other petroleum products encountered in the forensic laboratory, such as diesel fuel (18).

Acknowledgments

The authors would like to thank the ATF National Laboratory Center (ATF-NLC) for their support of this research project as well as the employees of the ATF-NLC for volunteering their time and automobiles for the study.

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