

# Standard Test Method for Extraction and Derivatization of Vegetable Oils and Fats from Fire Debris and Liquid Samples with Analysis by Gas Chromatography-Mass Spectrometry<sup>1</sup>

This standard is issued under the fixed designation E2881; the number immediately following the designation indicates the year of original adoption or, in the case of revision, the year of last revision. A number in parentheses indicates the year of last reapproval. A superscript epsilon  $(\varepsilon)$  indicates an editorial change since the last revision or reapproval.

ε<sup>1</sup> NOTE—Editorial corrections were made throughout in January 2015.

### 1. Scope

- 1.1 This test method covers the extraction, derivatization, and identification of fatty acids indicative of vegetable oils and fats in fire debris and liquid samples. This procedure will also extract animal oils and fats, as these are similar in chemical composition to vegetable oils and fats. Herein, the phrase "oils and fats" will be used to refer to both animal and vegetable derived oils and fats.
- 1.2 This test method is suitable for successfully extracting oil and fat residues having 8 to 24 carbon atoms.
- 1.3 The identification of a specific type of oil (for example, olive, corn, linseed) requires a quantitative analysis of the fatty acid esters and is beyond the scope of this test method.
- 1.4 This test method cannot replace the requisite knowledge, skills, or abilities acquired through appropriate education, training, and experience and should be used in conjunction with sound professional judgment.
- 1.5 The values stated in SI units are to be regarded as standard. No other units of measurement are included in this standard.
- 1.6 This standard does not purport to address all of the safety concerns, if any, associated with its use. It is the responsibility of the user of this standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use.

## 2. Referenced Documents

2.1 ASTM Standards:<sup>2</sup>

**E620** Practice for Reporting Opinions of Scientific or Technical Experts

- E1386 Practice for Separation of Ignitable Liquid Residues from Fire Debris Samples by Solvent Extraction
- E1388 Practice for Sampling of Headspace Vapors from Fire Debris Samples
- E1412 Practice for Separation of Ignitable Liquid Residues from Fire Debris Samples by Passive Headspace Concentration With Activated Charcoal
- E1413 Practice for Separation of Ignitable Liquid Residues from Fire Debris Samples by Dynamic Headspace Concentration
- E1492 Practice for Receiving, Documenting, Storing, and Retrieving Evidence in a Forensic Science Laboratory
- E1618 Test Method for Ignitable Liquid Residues in Extracts from Fire Debris Samples by Gas Chromatography-Mass Spectrometry
- E2154 Practice for Separation and Concentration of Ignitable Liquid Residues from Fire Debris Samples by Passive Headspace Concentration with Solid Phase Microextraction (SPME)
- E2451 Practice for Preserving Ignitable Liquids and Ignitable Liquid Residue Extracts from Fire Debris Samples

## 3. Summary of Test Method

- 3.1 If ignitable liquid analysis is required, it shall be performed prior to analysis for oils and fats as this test method is a destructive technique. A fire debris sample (or sub-sample) or an aliquot of a liquid is initially analyzed for ignitable liquid residues using standards listed in referenced documents.
- 3.2 The same sample of fire debris (or different sub-sample) or an additional aliquot of a liquid is then extracted with an organic solvent, and a derivatizing agent is added to convert either the free fatty acids and some triglycerides (for acid-catalyzed derivatization) or just the triglycerides (for base-catalyzed derivatization) to fatty acid methyl esters (FAMEs).
- 3.3 The organic layer of solvent is removed, filtered, and concentrated if necessary, using dry nitrogen, filtered air, or inert gas.
- 3.4 The derivatized extract is analyzed by gas chromatography-mass spectrometry (GC-MS).

<sup>&</sup>lt;sup>1</sup> This test method is under the jurisdiction of ASTM Committee E30 on Forensic Sciences and is the direct responsibility of Subcommittee E30.01 on Criminalistics. Current edition approved June 1, 2013. Published October 2013. DOI: 10.1520/F2881-13F01

<sup>&</sup>lt;sup>2</sup> For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.



3.5 Specific chemical components (fatty acid methyl esters) are identified by their retention times and mass spectra.

# 4. Significance and Use

- 4.1 This test method is useful when oils and fats are suspected as an ignition source or a fuel source in a fire.
- 4.1.1 The identification of oil and fat residues in samples from a fire scene can support the field investigator's opinion regarding the origin and cause of the fire.
- 4.1.2 The positive identification of fatty acid(s) does not necessarily mean that the fire was caused by self heating.
- 4.2 This test method specifically identifies fatty acid derivatives. Oils and fats are comprised primarily of triglycerides (which are fatty acids attached to a glycerol backbone), and some free fatty acids. Free fatty acids and triglycerides are not easily analyzed by the traditional ignitable liquid extraction techniques. Solvent extraction and derivatization to FAME will enable identification by GC-MS.
- 4.2.1 The identification of an individual fatty acid in fire debris samples does not confirm the presence of oils or fats; however, there are times when large quantities of the oil or fat may be extracted. In such cases a more positive identification can be made.
- 4.2.2 Oils and fats containing fatty acids with no double bonds will generally have no tendency to self-heat. With increasing unsaturation (1, 2 and 3 double bonds), the tendency to self-heat also increases, such that polyunsaturated fatty acids (PUFAs), such as C18:3, have a high tendency to self-heat.
- 4.3 This test method is a sensitive separation technique and can detect quantities as small as 3  $\mu L$  of oil or fat residue in an extract from a debris sample.
- 4.4 This test method shall be performed after all required traditional testing for ignitable liquid residues is completed.
- 4.5 This test method extracts liquids and residues from porous and nonporous materials of various sizes.
- 4.6 This test method can be hampered by coincident extraction of interfering compounds present in the fire debris samples.
- 4.7 This is a destructive technique and whenever possible the entire sample should not be used for the procedure. It is recommended that visual inspection be used to locate portions or areas exhibiting potential oily residue for sub-sampling which would preserve remaining portions for further analyses and also minimize solvent waste. The solvent extracted portions of the sample are not suitable for resampling.
- 4.8 Alternate methods of extraction, derivatization, or analysis exist and may be suitable for use in obtaining similar results and conclusions.
- 4.9 Biodiesel, an ignitable liquid, is a trans-esterified product containing FAMEs. The FAME compounds in biodiesel can be detected in fire debris using many fire debris extraction techniques followed directly by GC-MS analysis. Derivatization is not necessary to identify the FAMEs in biodiesel

## 5. Apparatus

- 5.1 Gas Chromatograph—A chromatograph capable of using capillary columns and being interfaced to a mass spectrometer
- 5.1.1 Sample Inlet System—A sample inlet system that can be operated in either split or splitless mode with capillary columns; the inlet system may use on-column technology.
- 5.1.2 *Column*—A capillary, bonded phase, methylsilicone or phenylmethylsilicone column or equivalent, or a polar capillary, bonded phase, such as a cyanopropyl-based fatty acid specific column, may be used to determine the presence of fatty acids.
- 5.1.2.1 A polar capillary, bonded phase, such as a cyanopropyl-based fatty acid specific column shall be used to perform comparative analysis between neat liquid samples, or fire debris samples, or both. Any column length or temperature program conditions may be used provided that each component of the reference mixture (see 6.8) is adequately separated on the polar column.
- 5.1.3 *GC Oven*—A column oven capable of reproducible temperature program operation in the range from 50 to 300°C.
- 5.2 *Mass Spectrometer*—Capable of acquiring mass spectra from m/z 40 to m/z 400 with unit resolution or better, with continuous data output.
- 5.2.1 Sensitivity and Resolution—The system shall be capable of detecting each component of the reference mixture (see 6.8) and providing sufficient ion intensity data to identify each component, either by computer library search or by comparison with reference spectra.
- 5.3 *Data Station*—A computerized data station capable of storing time sequenced mass spectral data from sample runs.
- 5.3.1 *Data Handling*—The data system shall be capable of performing, either through its operating system or by user programming, various data handling functions, including input and storage of sample data files, generation of extracted ion profiles, searching data files for selected compounds, and qualitative and semi-quantitative compound analysis.
- 5.3.2 Mass Spectral Libraries—The system shall be capable of retrieving a specified mass spectrum from a data file and comparing it against a library of mass spectra available to the data system. This capability is considered an aid to the analyst, who will use it in conjunction with chromatographic data and known reference materials to identify unknown components.
- 5.4 Syringes—A syringe capable of introducing a sample size in the range from 0.1 to  $10.0 \mu m$ .
- 5.5 *Steam bath*, for use in warming sample extracts in containers used during evaporation steps.

## 6. Reagents and Materials

6.1 *Purity of Reagents*—Reagent grade chemicals shall be used in all tests. It is intended that all reagents conform to the specifications of the Committee on Analytical Reagents of the American Chemical Society. Other grades may be used, provided it is first ascertained that the reagent is of sufficiently high purity to permit its use without lessening the accuracy of the determination.



- 6.2 *Solvent*—A suitable solvent, such as n-pentane, n-hexane, or n-heptane.
- 6.2.1 Solvent purity can be verified by evaporating to at least twice the extent used in the analysis and analyzing the evaporated solvent in accordance with Test Method E1618.
- 6.3 Derivatization Reagent—There are two types of derivatization processes: (1) acid-catalyzed, which will act on both triglycerides and free fatty acids; and (2) base-catalyzed, which only trans-esterify triglycerides. A suitable derivatization reagent, such as a 2N potassium hydroxide (KOH) in methanol solution (base-catalyzed) or a 10 % boron trifluoride in methanol solution (acid-catalyzed), will be chosen to convert the fatty acids and triglycerides to FAMEs.
- 6.4 Drying Agent—A suitable drying agent, such as anhydrous sodium sulfate.
- 6.5 Filter Apparatus, free of extractable hydrocarbons, oils, and fats.
- 6.6 Evaporation Accelerants—Compressed dry nitrogen, filtered air, or inert gas used in the concentration of solvent extracts.

- 6.7 Carrier Gas—Helium or hydrogen of purity 99.995 % or higher.
- 6.8 Reference Mixture—The reference mixture shall consist of a minimum of the following FAMEs: C16:0, C18:0, C18:1, C18:2, C18:3. Additional compounds may be included at the discretion of the analyst. The mixture should contain approximately equal parts by weight of the required fatty acids methyl esters in the chosen solvent or a traceable commercially available reference mixture. The final solution is prepared by diluting the above mixture such that the concentration of each component is no greater than 0.005 % weight/volume (0.05 micrograms/milliliter) in the chosen solvent. A typical chromatogram including components of the reference mixture on a typical chromatogram including components of the reference mixture on a fatty acid specific column is shown in Fig. 2.
- 6.9 Reference Oils and Fats—Oils and fats should be available for comparison and identification purposes.
- 6.9.1 Typically, FAMEs derived from reference oils and fats are diluted approximately 1:200 in an appropriate solvent and

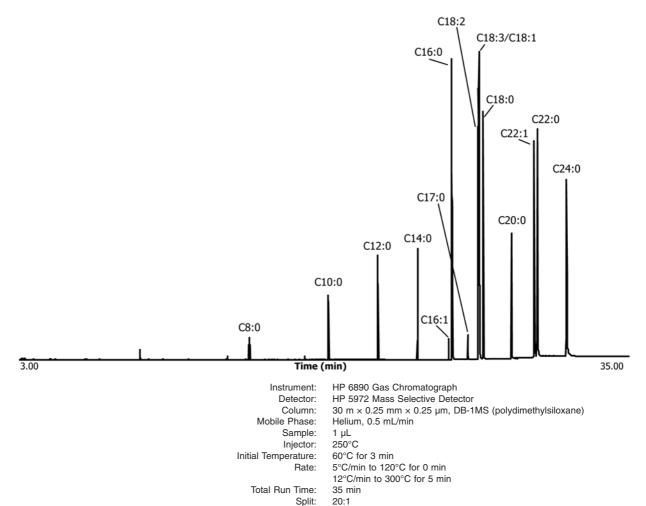
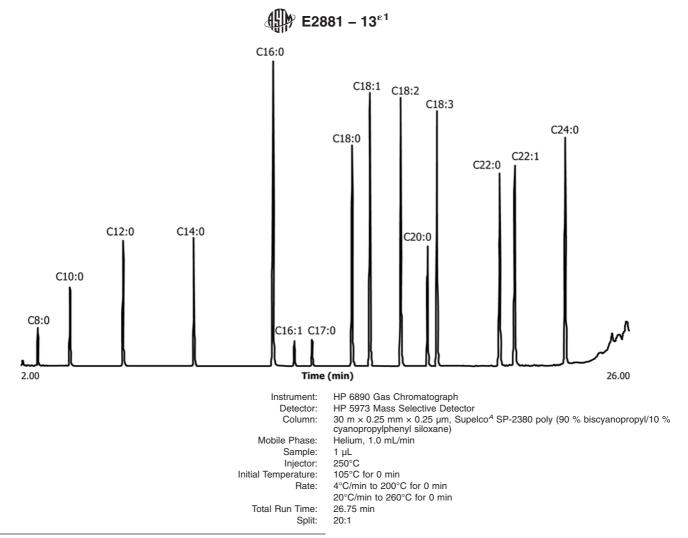


FIG. 1 Total Ion Chromatogram (TIC) of a FAME Reference Mixture on a DB-1MS Capillary Column Using Test Method E1618 GC-MS Conditions



<sup>&</sup>lt;sup>A</sup> A trademark by Sigma-Aldrich Co., LLC.

FIG. 2 Total Ion Chromatogram (TIC) of a FAME Reference Mixture on a SP-2380 Capillary Column Using a Program Optimized for Oil and Fat Analysis

derivatized using the same procedure that will be used on the debris and liquid samples. Depending on the column capacity and injection technique, derivatized oil and fat solutions can be concentrated to ensure detection of minor compounds.

6.10 *Glassware or Labware*—Clean glassware (beakers, test tubes, and vials) or disposable labware free of extractable hydrocarbons, oils, and waxes.

## 7. Equipment Calibration and Maintenance

- 7.1 Verify the consistent performance of the chromatographic instrument by using blanks and a known concentration of the reference mixture (see 6.8). Optimize gas flow periodically.
  - 7.2 Tune and Calibrate Mass Spectrometer:
- 7.2.1 Ensure proper operation of the mass spectrometer using perfluorotributylamine (PFTBA), or another appropriate calibration standard, according to the instrument manufacturer's specifications, prior to use. This should be done at least every day that the instrument is used or in accordance with manufacturer's recommendations.

- 7.2.2 Maintain tuning documentation as a portion of the quality control documentation.
  - 7.3 Equipment Maintenance:
- 7.3.1 Change septa and clean or replace injector liners on a periodic basis to avoid sample contamination by carryover of residual material from previous sample injections.

#### 8. Sample Handling Procedure

- 8.1 Only samples of appropriate dilution should be analyzed on a GC-MS system
- 8.2 Care must be taken to ensure that samples and extracts containing a small quantity of triglycerides and free fatty acids are not subjected to heat or evaporated to dryness prior to derivatization, as both of these may change the composition of the extract.
- 8.3 Analyze solvent blanks at least once each day that the instrument is used; maintain these analysis records. This will verify the purity of the solvent and potentially detect carryover or contamination.



- 8.4 Clean syringes thoroughly between injections to ensure no carryover.
- 8.4.1 Conduct carryover studies, and maintain records that demonstrate the adequacy of laboratory procedures to prevent carryover.
- 8.4.2 Running solvent blanks between each sample is recommended but not required if studies demonstrate that the cleaning procedure is adequate to prevent carryover.
- 8.5 Maintain reference files of known oils and fats that have been prepared and analyzed in the same manner as the questioned samples.

#### 9. Procedure

- 9.1 Observe the appropriate procedures for handling and documentation of all submitted samples as described in Practice E1412.
- 9.1.1 Open and examine the fire debris sample in order to determine that it is consistent with its description.
- 9.1.2 Resolve any discrepancies between the contributor's description of the evidence and the analyst's observations prior to the completion of the report
- 9.2 If appropriate or requested, perform analysis for ignitable liquid residues first. Methods for isolating ignitable liquid residues (ILR) from fire debris for analysis are described in Practices E1386, E1388, E1412, E1413, and E2154.
- 9.2.1 If available, use separate samples of the debris for ILR and FAME analyses because heating of the entire sample occurs during most ILR extraction practices which could accelerate the polymerization process of the triglycerides and free fatty acids, rendering them unavailable for the derivatization described in this test method
- 9.2.2 If Practice E1386 is to be used for ILR analysis, an aliquot of the solvent rinse from the FAMEs analysis can be obtained prior to derivatization, concentrated and analyzed using Test Method E1618.
- 9.3 Following ignitable liquid analysis, perform extraction for oils and fats. Determine which derivatization method is to be used. In either method, the system shall be dry (free of water) and have an excess of methanol present for the reaction to proceed to completion.
  - 9.3.1 Derivatization of Liquid Samples:
- 9.3.1.1 Dilute liquid in appropriate solvent (see 6.2) using at least 25-50  $\mu$ L (1 drop) of liquid to 10 mL of solvent. Use an equal amount of a reference oil sample as a positive control to process along with the questioned sample(s).
- 9.3.1.2 *Base-catalyzed derivatization*, add at least 0.5 mL of 2N KOH in methanol to the diluted liquid and vigorously mix (vortex mixing or shaken).
- 9.3.1.3 Acid-catalyzed derivatization, add at least 1.0 mL of a solution of 10 % boron trifluoride in methanol to the diluted liquid and seal. Heat at 60°C for 5 to 10 minutes in a closed container. Cool, add up to 1.0 mL of water, and vigorously mix (vortex mixing or shaken).
- 9.3.1.4 The layers should be allowed to separate (centrifuge if necessary), and the top (organic) layer removed. Filter the

- organic layer as needed with 0.45-µm filters (or finer) designed for organic extracts, such as polytetrafluoroethylene (PTFE) membrane filters.
- 9.3.1.5 Analyze the derivatized extract by GC-MS using the herein specified columns and appropriate conditions.
  - 9.3.2 Debris Samples:
- 9.3.2.1 Extract the selected portion of the sample in a clean container (free of extractable hydrocarbons, fats and oils) by adding sufficient solvent to thoroughly moisten the sample and mixing the solvent and debris for several minutes. Simple rinsing of nonporous surfaces may not adequately separate residues. Extracting the entire sample should be avoided whenever possible as this test is destructive.
  - 9.3.2.2 Remove the extract from the debris.
- Note 1—If water is present, after removal of the extract from the debris, allow the solvent and water to separate. Decant the solvent layer into a clean beaker and add a drying agent (for example, anhydrous sodium sulfate) to the sample extract until the drying agent flows freely without clumping. The amount of drying agent added will vary depending on the amount of water present. Before proceeding to the next step, remove the extract from the drying agent and transfer the extract to a new container.
- 9.3.2.3 Derivatize all or a portion of the extract (typically no more than 10 mL) by either the base-catalyzed or acid-catalyzed method as described above (see 9.3.1.2 or 9.3.1.3).
- 9.3.2.4 Use a reference oil sample (see 9.3.1.1) diluted in the same solvent as was used to extract the debris as a positive control to process along with the questioned extracted sample(s).
- 9.3.2.5 After derivatization, the layers should be allowed to separate (centrifuge if necessary), and the top (organic) layer removed. Filter the organic layer as needed with 0.45-µm filters (or finer) designed for organic extracts, such as polytetrafluoroethylene (PTFE) membrane filters.
- 9.3.2.6 Analyze the derivatized extract by GC-MS using the herein specified columns and appropriate conditions.
- 9.3.3 Extracts may be concentrated before or after derivatization. Place the extract in a chemical fume hood and evaporate at room temperature to a suitable final volume. Compressed dry nitrogen, filtered air, or an inert gas can be used to accelerate evaporation. Heat is not recommended for concentrating prior to derivatization as the oxidative polymerization of small quantities of extracted fatty acids and triglycerides could be accelerated by heat. Using heat to concentrate after derivatization is acceptable as polymerization does not occur with FAMEs.

## 10. Interpretation of Results

10.1 The identification of FAMEs in liquids and fire debris is performed using retention time and mass spectral data compared with a known standard, such as the reference mixture described in 6.8, or oil or fat reference material. It should be noted that the mere presence of fatty acids in fire debris does not confirm the presence of oils or fats; however, there are times when measurable quantities of the oil may be extracted. In such cases a more positive identification can be made.

- 10.2 Extracted Ion Profiling (EIP):
- 10.2.1 Extracted ion profiles may be used to elucidate fatty acid methyl ester peaks from other interferences in the chromatograms.
- 10.2.2 A data station is used to extract and draw extracted ion profiles (mass chromatograms) for major ions characteristic of fatty acid methyl esters.
- 10.2.3 Typically ions 67, 74, and 79 or the molecular weight ion for the fatty acid methyl ester of interest can be summed together to form a useful EIP. See Table 1 for a list of common fatty acids and the molecular weights of the resulting methyl esters
- 10.2.3.1 The ions 67 and 79 are indicators for methyl esters of linoleic acid (methyl octadecadienoate) and linolenic acid (methyl octadecatrienoate), which are acids known to be involved with self-heating oils.

# 11. Report Wording

- 11.1 Suggested wording for liquid, solid, and debris samples containing FAMEs:
  - 11.1.1 Fatty Acids Identified but no PUFAs Detected:
- 11.1.1.1 Oil (or fat) with very slight or low tendency toward spontaneous ignition was identified. This result does not eliminate the possibility that a substance with a higher tendency toward self-heating was present prior to the fire.
- 11.1.2 Fatty Acids Identified, but a Low Amount of Pufas Present (Small Amount of C18:2 and Even Less C18:3):
- 11.1.2.1 Oil (or fat) with a moderate tendency toward spontaneous ignition was identified. This result does not

- eliminate the possibility that a substance with a higher tendency toward self-heating was present prior to the fire.
- 11.1.3 Fatty Acids Identified, a Large Amount of PUFAs Identified:
- 11.1.3.1 Oil (or fat) with a high tendency toward spontaneous ignition was identified. This does not necessarily imply that spontaneous ignition occurred
- 11.2 Alternative Suggested Wording when FAMEs are Present in Liquid, Solid, and Debris Samples:—
- 11.2.1 Fatty acids were identified in Exhibit 1. Fatty acids are naturally occurring and are found in oils and fats. The fatty acids are consistent with known oils or fats; however, the original source could not be determined.
- 11.2.2 Fatty acids, which are typical components of oils or fats, were detected with a (no, low, moderate, or high) degree of unsaturation indicating a (no, low, moderate, or high) tendency of self-heating.
- 11.2.2.1 The above characterization of any oils or fats detected are appropriate only in cases in which the fatty acids are known to be unreacted.
- 11.2.3 In the above-mentioned wording, the term "fatty acids" may be specifically identified by their chemical name(s) or common name(s).
  - 11.3 Suggested Wording for Negative Reports:
  - 11.3.1 No Fatty Acids Identified:
- 11.3.1.1 No oil or fat was identified in the sample. This result does not preclude the possibility of oils or fats being consumed or polymerized in the fire.

TABLE 1 Common Fatty Acids and the Molecular Weights of the Resulting Methyl Esters†

Fatty Acid Designation	FAME Name	FAME Formula	FAME MW (amu)
C2:0	Methyl acetate	CH <sub>3</sub> COOCH <sub>3</sub>	74
C4:0	Methyl butyrate	C <sub>3</sub> H <sub>7</sub> COOCH <sub>3</sub>	102
C5:0	Methyl pentanoate	C <sub>4</sub> H <sub>9</sub> COOCH <sub>3</sub>	116
C6:0	Methyl hexanoate	C <sub>5</sub> H <sub>11</sub> COOCH <sub>3</sub>	130
C8:0	Methyl octanoate	C <sub>7</sub> H <sub>15</sub> COOCH <sub>3</sub>	158
C10:0	Methyl decanoate	C <sub>9</sub> H <sub>19</sub> COOCH <sub>3</sub>	186
C12:0	Methyl dodecanoate	C <sub>11</sub> H <sub>23</sub> COOCH <sub>3</sub>	214
C14:0	Methyl tetradecanoate	C <sub>13</sub> H <sub>27</sub> COOCH <sub>3</sub>	242
C16:0	Methyl hexadecanoate	C <sub>15</sub> H <sub>31</sub> COOCH <sub>3</sub>	270
C16:1(n-7)	Methyl hexadecenoate	C <sub>15</sub> H <sub>29</sub> COOCH <sub>3</sub>	268
C18:0	Methyl octadecanoate	C <sub>17</sub> H <sub>35</sub> COOCH <sub>3</sub>	298
C18:1(n-7)	Methyl octadecenoate	C <sub>17</sub> H <sub>33</sub> COOCH <sub>3</sub>	296
C18:1(n-9)	Methyl octadecenoate	C <sub>17</sub> H <sub>33</sub> COOCH <sub>3</sub>	296
C18:1(n-12)	Methyl octadecenaote	C <sub>17</sub> H <sub>33</sub> COOCH <sub>3</sub>	296
C18:2(n-6)	Methyl octadecadienoate	C <sub>17</sub> H <sub>31</sub> COOCH <sub>3</sub>	294
C18:3(n-3)	Methyl octadecatrienoate	C <sub>17</sub> H <sub>29</sub> COOCH <sub>3</sub>	292
C18:3(n-5)	Methyl octadecatrienoate	C <sub>17</sub> H <sub>29</sub> COOCH <sub>3</sub>	292
C18:3(n-6)	Methyl octadecatrienoate	C <sub>17</sub> H <sub>29</sub> COOCH <sub>3</sub>	292
C20:0	Methyl eicosanoate	C <sub>19</sub> H <sub>39</sub> COOCH <sub>3</sub>	326
C20:4(n-6)	Methyl eicosatetraenoate	C <sub>19</sub> H <sub>31</sub> COOCH <sub>3</sub>	318
C20:5(n-3)	Methyl eicosapentaenoate	C <sub>19</sub> H <sub>29</sub> COOCH <sub>3</sub>	316
C22:0	Methyl docosanoate	C <sub>21</sub> H <sub>43</sub> COOCH <sub>3</sub>	354
C22:1(n-9)	Methyl docosenoate	C <sub>21</sub> H <sub>41</sub> COOCH <sub>3</sub>	352
C22:6(n-3)	Methyl docosahexaenoate	C <sub>21</sub> H <sub>31</sub> COOCH <sub>3</sub>	342
C24:0	Methyl tetracosanoate	C <sub>23</sub> H <sub>47</sub> COOCH <sub>3</sub>	382
C24:1(n-9)	Methyl tetracosenoate	C <sub>23</sub> H <sub>45</sub> COOCH <sub>3</sub>	380
C26:0	Methyl hexacosanoate	C <sub>25</sub> H <sub>51</sub> COOCH <sub>3</sub>	410

<sup>†</sup> Editorially corrected.



- 11.4 Limitations:
- 11.4.1 It should be noted that this is a qualitative technique; amounts are relative and are based upon the analyst's experience and judgment.
- 11.4.2 The identification of a specific type of oil requires a quantitative analysis of the fatty acid esters and is beyond the scope of this test method.
- 11.5 Refer to Practice E620 and Test Method E1618, Section 12, for general information on report writing.

#### 12. Precision and Bias

12.1 This test method is qualitative, and therefore precision and bias cannot be measured.

## 13. Keywords

13.1 animal fats; fatty acid methyl esters; fire debris samples; forensic sciences; gas chromatography; ignitable liquid residues; mass spectrometry; vegetable oils

#### **ANNEX**

(Mandatory Information)

### A1. SAMPLE STORAGE

- A1.1 After extraction, store the original debris or liquid sample using appropriate procedures for handling and documentation as set forth in Practice E1492.
- A1.1.1 Extract Storage Short-Term—Extracts prior to derivatization should be stored in a refrigerator in a sealed vial or container (free of extractable hydrocarbons) to inhibit further polymerization and prevent evaporation
- A1.1.1.1 Extracts after derivatization can be evaporated to dryness at room temperature and stored in a sealed vial or container (free of extractable hydrocarbons) at room temperature.
- A1.1.2 Extract Storage Long-Term—Only derivatized extracts should be stored long-term. The procedures set forth in Practice E2451 should be followed for long-term storage of extracts or liquid samples.
- A1.1.3 *Storage of ILR Extracts*—Follow the procedures set forth in Practice E2451 for storage of ILR extracts.

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