Standard Test Method for Determination of Ethanol and Methanol Content in Fuels Containing Greater than 20% Ethanol by Gas Chromatography[1]

This standard is issued under the fixed designation D5501; 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 (ε) indicates an editorial change since the last revision or reapproval.

1. Scope

1.1 This test method covers the determination of the ethanol content of hydrocarbon blends containing greater than 20 % ethanol. This method is applicable to denatured fuel ethanol, ethanol fuel blends, and mid-level ethanol blends.

1.1.1 Ethanol is determined from 20 % by mass to 100 % by mass and methanol is determined from 0.01 % by mass to 0.6 % by mass. Equations used to convert these individual alcohols from percent by mass to percent by volume are provided.

NOTE 1—Fuels containing less than 20 % ethanol may be quantified using Test Method D5599, and less than 12 % ethanol may be quantified using Test Method D4815.

1.2 This test method does not purport to identify all individual components common to ethanol production or those components that make up the denaturant or hydrocarbon constituent of the fuel.

1.3 Water cannot be determined by this test method and shall be measured by a procedure such as Test Method D1364 and the result used to correct the concentrations determined by this method.

1.4 This test method is inappropriate for impurities that boil at temperatures higher than 225 °C or for impurities that cause poor or no response in a flame ionization detector, such as water.

1.5 The values stated in SI units are to be regarded as the standard. The values given in parentheses are provided for information purposes only.

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:*[2]

D1298 Test Method for Density, Relative Density, or API Gravity of Crude Petroleum and Liquid Petroleum Products by Hydrometer Method

D1364 Test Method for Water in Volatile Solvents (Karl Fischer Reagent Titration Method)

D4052 Test Method for Density, Relative Density, and API Gravity of Liquids by Digital Density Meter

D4057 Practice for Manual Sampling of Petroleum and Petroleum Products

D4175 Terminology Relating to Petroleum Products, Liquid Fuels, and Lubricants

D4307 Practice for Preparation of Liquid Blends for Use as Analytical Standards

D4626 Practice for Calculation of Gas Chromatographic Response Factors

D4806 Specification for Denatured Fuel Ethanol for Blending with Gasolines for Use as Automotive Spark-Ignition Engine Fuel

D4815 Test Method for Determination of MTBE, ETBE, TAME, DIPE, tertiary-Amyl Alcohol and C_1 to C_4 Alcohols in Gasoline by Gas Chromatography

D5599 Test Method for Determination of Oxygenates in Gasoline by Gas Chromatography and Oxygen Selective Flame Ionization Detection

D5798 Specification for Ethanol Fuel Blends for Flexible-Fuel Automotive Spark-Ignition Engines

D6299 Practice for Applying Statistical Quality Assurance and Control Charting Techniques to Evaluate Analytical Measurement System Performance

D6792 Practice for Quality System in Petroleum Products and Lubricants Testing Laboratories

E203 Test Method for Water Using Volumetric Karl Fischer Titration

---

[1] This test method is under the jurisdiction of ASTM Committee D02 on Petroleum Products, Liquid Fuels, and Lubricants and is the direct responsibility of Subcommittee D02.04.0L on Hydrocarbon Analysis.

Current edition approved Oct. 1, 2016. Published November 2016. Originally approved in 1994. Last previous edition approved in 2012 as D5501 – $12^{\varepsilon1}$. DOI: 10.1520/D5501-12R16.

[2] 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.

Copyright © ASTM International, 100 Barr Harbor Drive, PO Box C700, West Conshohocken, PA 19428-2959. United States

E355 Practice for Gas Chromatography Terms and Relationships

E594 Practice for Testing Flame Ionization Detectors Used in Gas or Supercritical Fluid Chromatography

E1064 Test Method for Water in Organic Liquids by Coulometric Karl Fischer Titration

E1510 Practice for Installing Fused Silica Open Tubular Capillary Columns in Gas Chromatographs

3. Terminology

3.1 *Definitions*—This test method makes reference to many common chromatographic procedures, terms, and relationships. Detailed definitions can be found in Terminology D4175, and Practices E355 and E594.

3.2 *Definitions:*

3.2.1 *mass response factor (MRF), n*—constant of proportionality that converts area to mass percent.

3.2.2 *relative mass response factor (RMRF), n*—mass response factor of a component divided by that of another component.

3.2.2.1 *Discussion*—In this test method, the mass response factors are relative to that of n-heptane.

3.2.3 *tangential skimming, n—in gas chromatography*, integration technique used when a "rider" peak elutes on the tail of a primary peak.

3.2.3.1 *Discussion*—Since the majority of the area beneath the rider peak belongs to the primary peak, in tangential skimming the top of the primary peak tail is used as the baseline of the rider peak, and the triangulated area beneath the rider peak is added to the primary peak.

3.3 *Abbreviations:*

3.3.1 *MRF*—mass response factor

3.3.2 *RMRF*—relative mass response factor

4. Summary of Test Method

4.1 A representative aliquot of the fuel ethanol sample is introduced into a gas chromatograph equipped with a polydimethylsiloxane bonded phase capillary column. Carrier gas transports the vaporized aliquot through the column where the components are chromatographically separated in order of boiling point temperature. Components are sensed by a flame ionization detector as they elute from the column. The detector signal is processed by an electronic data acquisition system. The ethanol and methanol components are identified by comparing their retention times to the ones identified by analyzing standards under identical conditions. The concentrations of all components are determined in mass percent by normalization of the peak areas. After correction for water content, results may be reported in mass percent or volume percent.

5. Significance and Use

5.1 This test method provides a method of determining the percentage of ethanol in an ethanol-gasoline fuel blend over the range of 20 % by mass to 100 % by mass for compliance with fuel specifications and federal or local fuel regulations.

5.2 Ethanol content of denatured fuel ethanol for gasoline blending is required in accordance with Specification D4806.

TABLE 1 Typical Operating Conditions

Column Temperature Program		
Column length	100 m	150 m
Initial temperature	15 °C	60 °C
Initial hold time	12 min	15 min
Program rate	30 °C ⁄min	30 °C ⁄min
Final temperature	250 °C	250 °C
Final hold time	19 min	23 min
Injector		
Temperature	300 °C	
Split ratio	200:1	
Sample size	0.1 µL to 0.5 µL	
Detector		
Type	Flame ionization	
Temperature	300 °C	
Fuel gas	Hydrogen (30 mL ⁄min)	
Oxidizing gas	Air (300 mL/min)	
Make-up gas	Helium or Nitrogen (30 mL/min)	
Date rate	20 Hz	
Carrier Gas		
Type	Helium or Hydrogen[A]	
Average linear velocity	21 cm ⁄s to 24 cm/s (constant flow)	

[A] Use of hydrogen carrier gas requires additional safety considerations.

5.3 Ethanol content of ethanol fuel blends for flexible-fuel automotive spark-ignition engines is required in accordance with Specification D5798.

6. Apparatus

6.1 *Gas Chromatograph,* capable of operating at the conditions listed in Table 1. A heated flash vaporizing injector designed to provide a linear sample split injection (for example, 200:1) is required for proper sample introduction. Carrier gas controls shall be of adequate precision to provide reproducible column flows and split ratios in order to maintain analytical integrity. Pressure and flow control devices shall be designed to attain the linear velocity required in the column used. A hydrogen flame ionization detector with associated gas controls and electronics, designed for optimum response with open tubular columns, is required.

6.2 *Sample Introduction*—Automatic liquid syringe sample injection to the splitting injector. Devices capable of 0.1 µL to 0.5 µL injections are suitable.

NOTE 2—Inadequate splitter, poor injection technique, and overloading the column can result in poor resolution. Avoid overloading, particularly of the ethanol peak, and eliminate this condition during analysis.

6.3 *Column*—The precision for this test method was developed utilizing a fused silica open tubular column with non-polar polydimethylsiloxane bonded (cross-linked) phase internal coating. Any column with equivalent or better chromatographic efficiency, resolution, and selectivity to those described in 6.3.1 may be used.

6.3.1 Open tubular column with a non-polar polydimethylsiloxane bonded (cross-linked) phase internal coating, either 150 m by 0.25 mm with a 1.0 µm film thickness, or 100 m by 0.25 mm with a 0.5 film thickness have been found suitable. The 150 m column is recommended due to its higher resolution. Follow Practice E1510 for column installation.

6.4 *Electronic Data Acquisition System*—Any data acquisition and integration device used for quantification of these analyses must meet or exceed these minimum requirements:

6.4.1 Capacity for at least 80 peaks/analysis,

6.4.2 Normalized percent calculation based on peak area and using response factors,

6.4.3 Identification of individual components based on retention time,

6.4.4 Noise and spike rejection capability,

6.4.5 Sampling rate for narrow (<1 s) peaks,

6.4.6 Positive and negative sloping baseline correction,

6.4.7 Peak detection sensitivity compensation for narrow and broad peaks, and

6.4.8 Capable of integrating non-resolved peaks by perpendicular drop or tangential skimming as needed.

7. Reagents and Materials

7.1 *Purity of Reagents*—Reagent grade chemicals shall be used in all tests. Unless otherwise indicated, it is intended that all reagents conform to the specifications of the Committee on Analytical Reagents of the American Chemical Society where such specifications are available.[3]

7.2 *Carrier Gas,* helium or hydrogen, with a minimum purity of 99.95 mol%. Oxygen removal systems and gas purifiers should be used. (**Warning—**Compressed gas under high pressure.) Use of hydrogen carrier gas may require additional safety considerations.

7.3 *Detector Gases,* hydrogen, air, and nitrogen. The minimum purity of the gases used should be 99.95 % for the hydrogen and nitrogen. The air should be hydrocarbon-free grade. Gas purifiers are recommended for the detector gases. (**Warning—**Hydrogen, extremely flammable gas under high pressure.) (**Warning—**Air and nitrogen, compressed gases under high pressure.)

7.4 *Standards for Calibration and Identification*—Standards of all components to be analyzed are required for establishing identification by retention time as well as calibration for quantitative measurements. These materials shall be of known purity and free of the other components to be analyzed.

7.4.1 To verify the purity of blend components, analyze each compound by the same technique for which the blend is intended or by another suitable technique.

7.4.2 Check for other impurities such as water. Water cannot be determined with sufficient accuracy by most GC methods and shall be measured by other procedures such as Test Method D1364, or equivalent, and the result used to normalize the chromatographic value. If any of the impurities found are components present in the blend, determine their concentrations and make appropriate corrections.

7.4.3 *Ethanol*—Absolute ethanol, 99.5 minimum mass percent. Methanol content should be less than 0.01 mass percent. (**Warning—**Flammable and may be harmful or fatal if ingested or inhaled.)

7.4.4 *Methanol*—(**Warning—**Flammable and may be harmful or fatal, if ingested or inhaled.) Minimum purity of 99 mass percent, and free of ethanol.

7.4.5 *Heptane*—(**Warning—**Flammable and may be harmful or fatal, if ingested or inhaled.)

7.4.6 *Hydrocarbon Diluent*—n-Octane or isooctane, used for preparation of calibration standards. The diluent shall contain <0.01 % of heptane, methanol, or ethanol. (**Warning—**Flammable and may be harmful or fatal if ingested or inhaled.)

7.5 *Linearity Mixture*—A mixture of known composition containing 10-20 hydrocarbons, ranging from C5 to C11, used for split injector linearity testing. (**Warning—**Flammable and may be harmful or fatal if ingested or inhaled.)

8. Sampling

8.1 Denatured ethanol may be sampled into an open container since a vapor pressure of less than 21 kPa (3 psi) is expected. Refer to Practice D4057 for instruction on manual sampling from bulk storage into open containers. Stopper the container immediately after drawing the sample.

8.2 To minimize loss of hydrocarbon light ends, chill the sample before transferring an aliquot into an auto sampler vial. Seal immediately. Obtain the test sample for analysis directly from the sealed auto sampler vial.

9. Preparation and Verification of Apparatus

9.1 Install and condition the column in accordance with the manufacturer's or supplier's instructions. After conditioning, attach column outlet to flame ionization detector inlet and check for leaks throughout the system. When leaks are found, tighten or replace fittings before proceeding.

9.2 Adjust the carrier gas flow rate so that the average linear gas velocity, at the initial temperature of the run, is between 21 cm/s and 24 cm/s, as determined by Eq 1. Flow rate adjustment is made by raising or lowering the carrier gas pressure (head pressure) to the injector. Maintain constant flow throughout the analysis.

$$\bar{\mu} = \frac{L}{t_m} \quad (1)$$

where:

$\bar{\mu}$ = average linear gas velocity (cm/s),
L = column length (cm), and
t_m = retention time of methane.

9.3 Adjust the operating conditions of the gas chromatograph (Table 1) and allow the system to equilibrate.

9.4 *Linearity*—The linearity of the gas chromatograph system shall be established prior to the analysis of samples.

9.4.1 The optimal split ratio is dependent upon the split linearity characteristics of the particular injector and the sample capacity of the column. The capacity of a particular column for a sample component is dependent on the amount and polarity of the liquid phase (loading or film thickness) and the ratio of the column temperature to the component boiling point (vapor pressure). Overloading of the column can cause loss of resolution for some components and, since overloaded peaks are skewed, variance in retention times. This can lead to

---

[3] *Reagent Chemicals, American Chemical Society Specifications*, American Chemical Society, Washington, DC. For Suggestions on the testing of reagents not listed by the American Chemical Society, see *Annual Standards for Laboratory Chemicals,* BDH Ltd., Poole, Dorset, U.K., and the *United States Pharmacopeia and National Formulary,* U.S. Pharmacopeial Convention, Inc. (USPC), Rockville, MD.

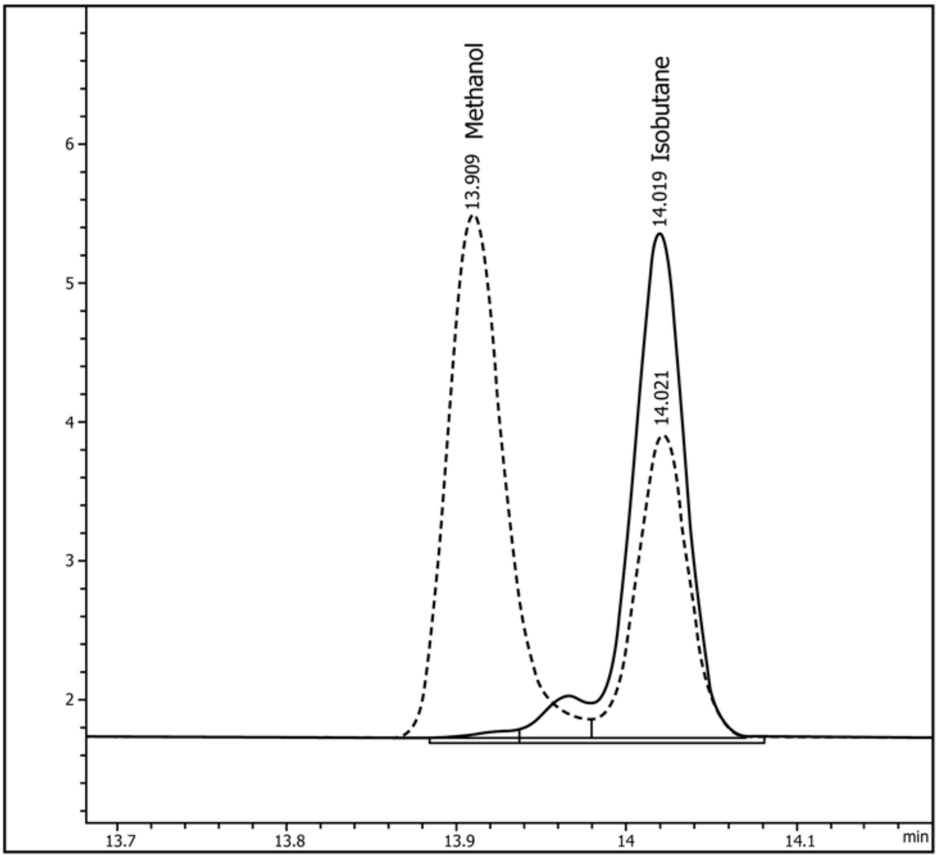


FIG. 1 Chromatogram Overlay Showing Possible Methanol Interferences on 150 m Column

erroneous component identification. During column evaluations and split linearity studies, be aware of any peaks that appear front skewed, indicating column overload. Note the component size and avoid conditions leading to this problem during actual analysis. Refer to Practice E594 for further guidance.

9.4.2 The injector split linearity is dependent on the nature of the compound (boiling point, molecular weight, etc.) splitter and liner design, injection volume, and the linear velocity of sample through the inlet. Establish the splitting injector linearity to determine the proper quantitative parameters and limits. Analyze the linearity mixture, 7.5, following the gas chromatographic analysis procedure in Section 12. Calculate the normalized mass percent (using Eq 8, Eq 5, and Eq 6) of each component using a relative mass response factor of 1 for all hydrocarbon compounds. The determined mass percent for each component shall match the gravimetric known concentration within ±3 % relative.

9.4.3 Verify the linearity of the flame ionization detector (FID). Refer to Practice E594 for a suggested procedure. A plot of the peak areas versus concentration for prepared methanol or ethanol standards in the concentration range of interest should be linear. If the plot is not linear or does not conform to these requirements then adjust the split ratio.

9.5 *Resolution and Integration:*

9.5.1 Optimize the system to resolve methanol from isobutane on the 150 m column using the conditions in this method. With higher amounts of isobutane, the possibility for coelution increases. Improved resolution has been achieved by optimizing the linear velocity to 24 cm⁄s. See Fig. 1.

9.5.2 Methanol might not be completely resolved from Butene-1/Isobutylene on the 100 m column using the conditions in this method. Use tangential skimming to properly integrate the methanol.

9.5.3 Ethanol might not be completely resolved from 3-methyl-1-butene. If so, use tangential skimming to integrate the ethanol. See Fig. 2.

9.5.4 Set integration parameters such that any component of at least 0.002 % by mass is integrated.

9.6 *Peak Shape:*

9.6.1 System overload of ethanol can be determined upon visual inspection of the peak. A flat top indicates electronic saturation. A wide "fin" shape can indicate column overload. In either case, investigate and make adjustments.

9.6.2 Hydrocarbon peaks shall be symmetrical. Slight tailing of polar compounds (methanol, ethanol) is typical, but excessive tailing should be investigated. Verify that the inlet liners are thoroughly deactivated and the column shows appropriate deactivation towards methanol and ethanol. Fig. 3 shows an example of poor chromatography.

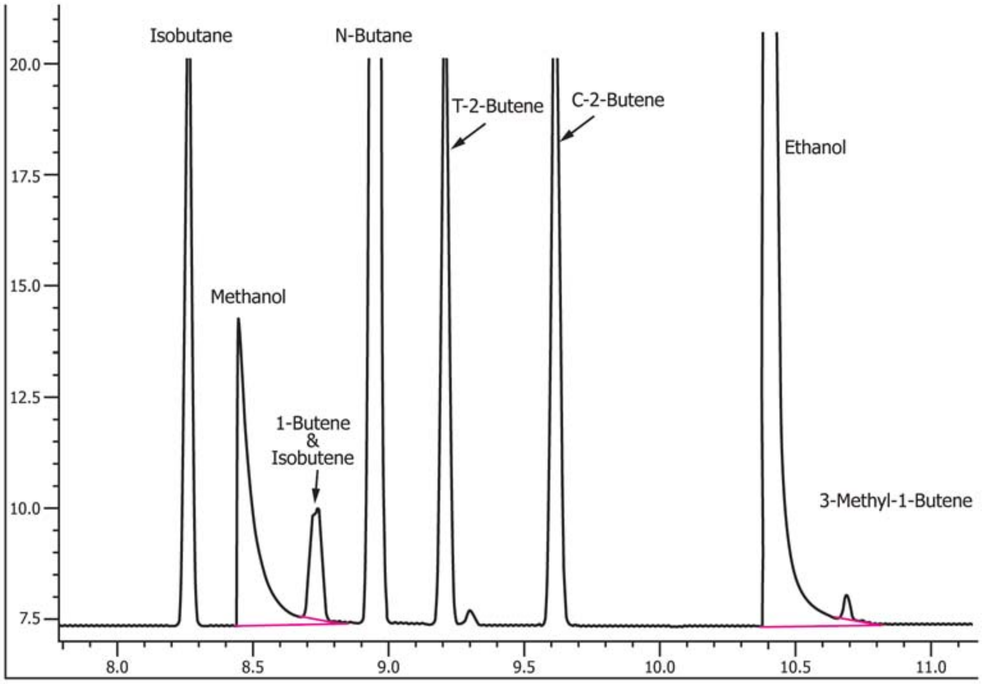
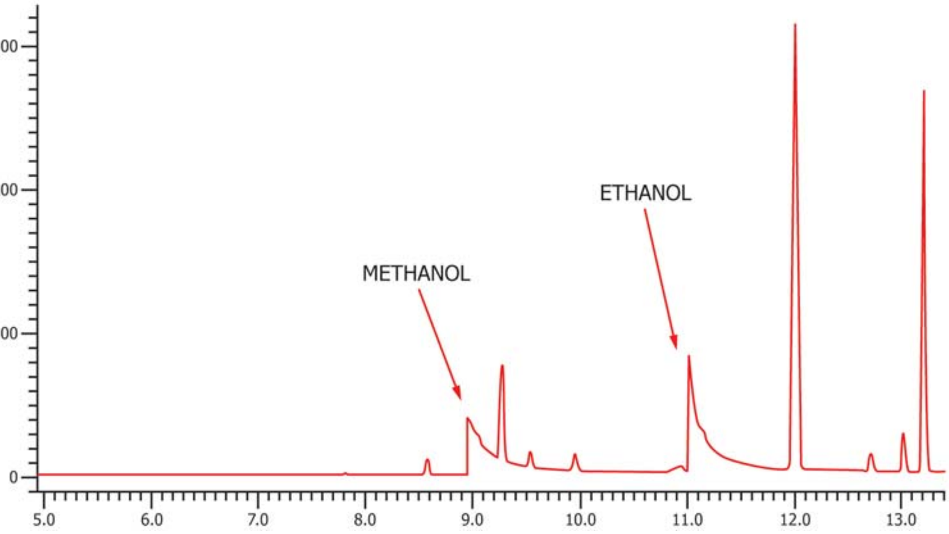


FIG. 2 Integration of Methanol and Ethanol on 100 m Column Using Tangential Skimming



NOTE 1—Excessive tailing and delayed elution may result from adsorption, dead volume, or damaged column.

FIG. 3 Example of Poor Chromatography

10. Calibration and Standardization

10.1 *Identification*—Determine the retention time of ethanol and methanol by injecting amounts of each, either separately or in known mixtures, in concentrations expected in the final blend.

10.2 *Calibration Standards:*

10.2.1 Determine the purity of the oxygenate reagents and make corrections for the impurities found. Whenever possible, use stock chemicals of at least 99.5 % purity. Correct the purity of the components for water content, determined by Test Method D1364 or equivalent. Example calculations are presented in Appendix X1.

10.2.2 Gravimetrically prepare standards that are blended according to Practice D4307. Chill components prior to blending. Prepare standards that cover the expected range of ethanol and methanol in a hydrocarbon diluent (n-octane or isooctane) and containing a known amount of heptane. Examples of standards covering the range of 20 % by mass to 99 % by mass ethanol are given in Table 2.

10.3 *Calibration Linearity*—Analyze the standards from 10.2 according to the procedure in Section 12. An example chromatogram is found in Fig. 4. For ethanol, methanol, and n-heptane, the plot of the peak areas versus concentration shall be linear with a minimum r^2 of 0.995. Do not force the ethanol

TABLE 2 Recommended Matrix of Calibration Standards Ranging from 20 % to 99 % by Mass

	Mix 1	Mix 2	Mix 3	Mix 4	Mix 5
Methanol, % by mass	0.6	0.5	0.3	0.2	0.1
Ethanol, % by mass	20.0	50.0	75.0	90.0	99.4
Heptane, % by mass	10.0	10.0	10.0	4.0	0.5
Hydrocarbon diluent,[A] % by mass	69.4	39.5	14.8	5.8	0.0

[A] Hydrocarbon diluent is free of heptane and any other compounds that would interfere with the calibration. See 7.4.

calibration through the origin (zero). For ethanol, a peak area of zero shall correspond to an ethanol concentration between –3 % and +3 % by mass (see Note 3). If the plot does not conform to these requirements, troubleshoot the system and repeat the analyses using fresh vials of standard until requirements are met. Typical troubleshooting measures include increasing the split ratio, reducing the injection amount, correcting the integration of the ethanol and methanol peaks, and checking for active sites in column and inlet liner. Example linearity plots for ethanol are given in Fig. 5.

10.4 *Calibration*—Calibration techniques may be classified as absolute or relative, and are typically dictated by the chromatographic software. Only after linearity is established, calibrate with one of the techniques below.

10.4.1 *Absolute Calibration*—Establish the correction factor or equation that directly relates the peak area to the component concentration. If available, multi-point calibration is preferred.

10.4.1.1 *Single-point*—Calculate the mass response factor (MRF) of ethanol in each calibration standard using Eq 2 according to Practice D4626. The average response factor determine at each concentration shall be used in the calibration. Repeat for methanol and heptane. Assign the heptane response factor to any unknowns.

$$\mathrm{MRF}_{(i)} = \mathrm{mass\%}_{(i)}/\mathrm{area}_{(i)} \qquad (2)$$

where:

$MRF_{(i)}$ = the mass response factor of component i,
$Mass\%_{(i)}$ = the mass percent of component i, and
$Area_{(i)}$ = the peak area of component i.

10.4.1.2 *Multi-point*—With this technique, calibration and linearity verification may be completed in the same step. Analyze the standards prepared in 10.2, according to the procedure in Section 12. Generate individual calibration curves for methanol, ethanol, and heptane following the software manufacturer's instructions. Calibration curves shall be linear with a minimum r^2 of 0.995. Verify that the calibration curve of ethanol passes near the origin (see Note 3), but do not force the origin. Force methanol and heptane calibrations through zero if necessary to quantify small peaks. Failure to quantify small unknown peaks will result in incorrect ethanol determination. The calibration equation of heptane shall be assigned to all unknowns.

NOTE 3—Software packages can differ in designation of the axes. If the peak area is on the x-axis, the y-intercept of the ethanol curve shall be between –3 % and +3 % by mass. If the peak area is on the y-axis, solving the calibration equation for y = 0 shall give a result between –3 % and +3 % by mass.

10.4.2 *Relative Calibration*—Tabulate the mass response factors (MRF) of ethanol, methanol, and n-heptane according to Eq 2. Then calculate the relative mass response factor (RMRF) of methanol and ethanol relative to heptane with Eq 3. Typical relative mass response factors for the components of interest are found in Table 3.

$$\mathrm{RMRF}_{(i)} = \mathrm{MRF}_{(i)}/\mathrm{MRF}_{(\mathrm{heptane})} \qquad (3)$$

where:

$RMRF_{(i)}$ = the relative mass response factor of component i relative to n-C_7,
$MRF_{(i)}$ = the mass response factor of component i, and
$MRF_{(heptane)}$ = the mass response factor of heptane.

Average the experimental ethanol relative mass response factors determined for each standard. Use the average RMRF as the calibration value. Repeat for methanol. Assign heptane and all unknowns an RMRF of 1.000.

10.5 After completing the calibration, calculate the composition of each calibration standard according to the procedure in Section 13. Verify that the normalized results are in agreement with theoretical ethanol values within ±0.5 % and with theoretical methanol values within ±0.05 %.

11. Quality Control

11.1 Conduct a regular statistical quality assurance (quality control) program, monitoring both precision and accuracy, in accordance with the techniques of Practice D6299 or equivalent. Measure the ethanol and methanol concentrations using the procedure outlined in Section 12. Confirm the performance of the instrument or the test procedure after each calibration and on each day of use thereafter. Include at least one quality control sample of known ethanol and methanol content.

11.1.1 Standard(s) of known concentration may be supplied from a vendor, cross-check program, or may be prepared gravimetrically according to X1.1. If possible, use of a certified reference material is recommended. Test at least one standard for each class of ethanol routinely analyzed (such as denatured fuel ethanol (>95 % Ethanol), ethanol fuel blends (51 % to 83 % ethanol), and mid-level ethanol blends (20 % to 51 % ethanol).

11.1.2 Prepare standard(s) in sufficient volume to allow for a minimum of 30 quality control measurements to be made on one batch of material. Properly package and store the quality control samples to ensure that all analyses of quality control samples from a given lot are performed on essentially identical material. Use of the Q-procedure in Practice D6299 is recommended when switching between batches of control sample.

12. Gas Chromatographic Analysis Procedure

12.1 Set the instrument operating variables. See Table 1 for typical operating conditions.

12.2 Set instrumental sensitivity and integration parameters such that any component of at least 0.002 mass % is detected and integrated.

12.3 Inject 0.1 µL to 0.5 µL of sample into the injection port and start the analysis. Obtain a chromatogram and verify the integration (see 9.5) and peak identification. Generate the peak

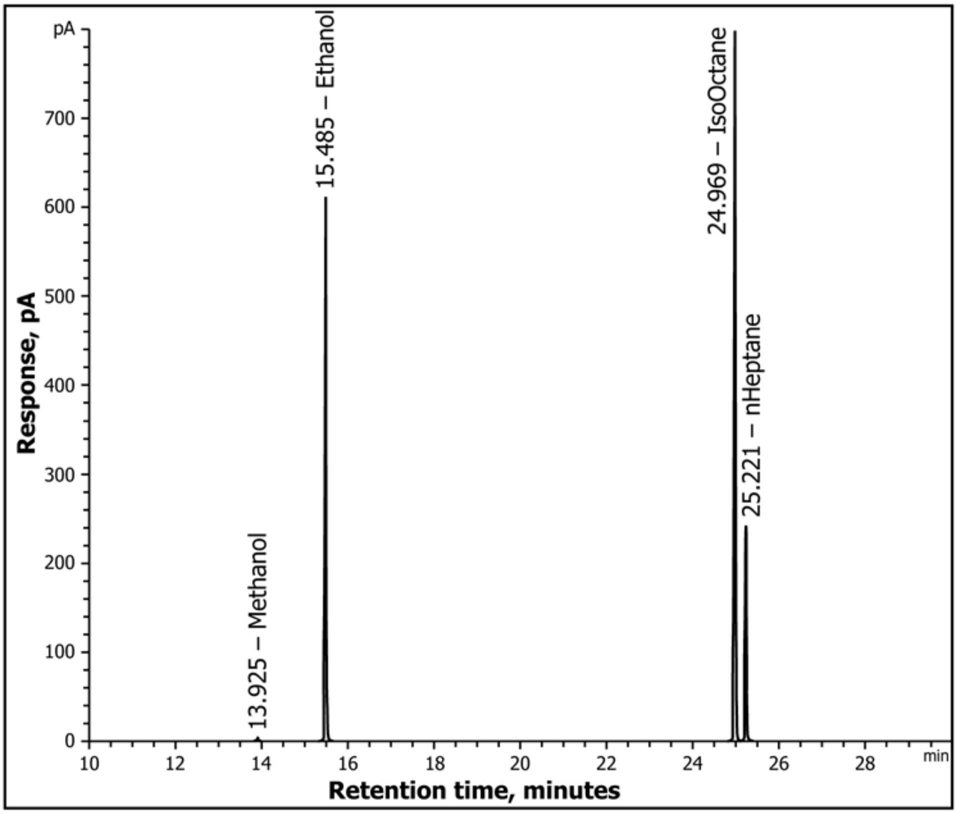


FIG. 4 Sample Chromatogram of Calibration Mixture on 150 m Column

integration or system report. Quantify results using calculations in Section 13. Sample chromatograms are shown in Fig. 6 and Fig. 7.

13. Calculation

13.1 Apply the appropriate calibration factor determined in 10.4 to each peak area.

13.1.1 For single point or relative mass response factor calibration, calculate the response corrected peak area (AR_i) of each component according to Eq 4. Repeat for each peak, using response factors determined for individual compounds and using the heptane calibration for unknowns.

$$AR_i = area_i \times RF(F)_i \quad (4)$$

where:

AR_i = response corrected peak area (i),
$area_i$ = peak area of component (i), and
$RF(F)_i$ = mass response factor (MRF) or relative mass response factor (RMRF) of component (i) relative to n-C_7.

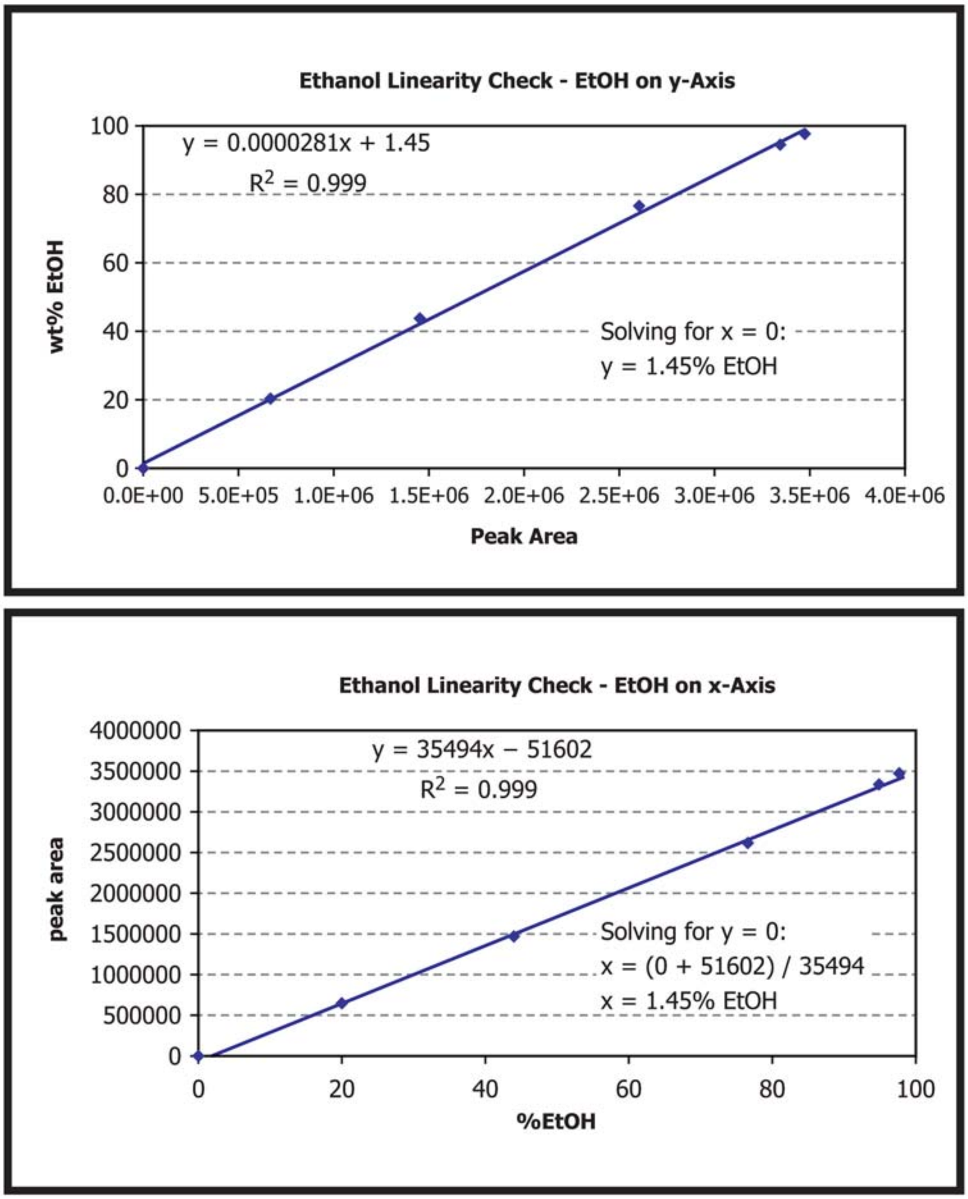
13.1.2 For multi-point calibration, input the peak area into the corresponding calibration equation determined in 10.4.1.2. Solve for the raw mass percent of component i, which is equivalent to AR_i. Repeat for each peak, using calibration equations determined for individual compounds and using the heptane calibration equation for unknowns.

13.1.3 When using absolute calibration (single- or multi-point) it is possible to monitor mass percent recovery. Calculate the raw mass percent recovery of the sample according to Eq 5. When properly calibrated, a fully eluting sample is expected to give a raw recovery of 95 % to 105 % by mass. Failure to meet these limits can indicate a poor calibration and/or integration, high water content, or a change in the system since the time of calibration.

$$\text{Mass\%} \quad \text{recovery} = AR_t \quad (5)$$

where:

AR_t = sum of AR_i for all detected peaks.



NOTE 1—The regression is linear with a correlation coefficient greater than 0.995 and the calibration curve passes within 3 % by mass ethanol of the origin.

FIG. 5 Calibration Linearity Checks are Shown Using Either Axis for Ethanol Content

TABLE 3 Pertinent Component Data

	Typical Mass Relative Response Factors[A]	Density at 20 °C, g/mL	Relative Density at 15.56 °C
Methanol	3.20	0.791	0.796
Ethanol	2.06	0.789	0.794

[A] where *n*-heptane = 1.

13.2 Determine the normalized relative mass percent of the individual alcohols by using the following equation:

$$RM_i = \frac{AR_i \times 100}{AR_t} \tag{6}$$

where:

RM_i = normalized relative percent by mass of the individual alcohols,

AR_i = response corrected peak area of component i, and

AR_t = sum of AR_i for all detected peaks.

13.3 Obtain the percent by mass of water in the sample. Test Methods D1364, E203, E1064, or equivalent, may be used.

13.4 Determine the percent by mass of the alcohols of interest by using the following equation:

$$M_i = \frac{RM_i \times (100 - \text{mass \% water in sample})}{100} \tag{7}$$

where:

M_i = percent by mass of the individual alcohol being determined, and

RM_i = normalized relative percent by mass of the individual alcohol from Eq 6.

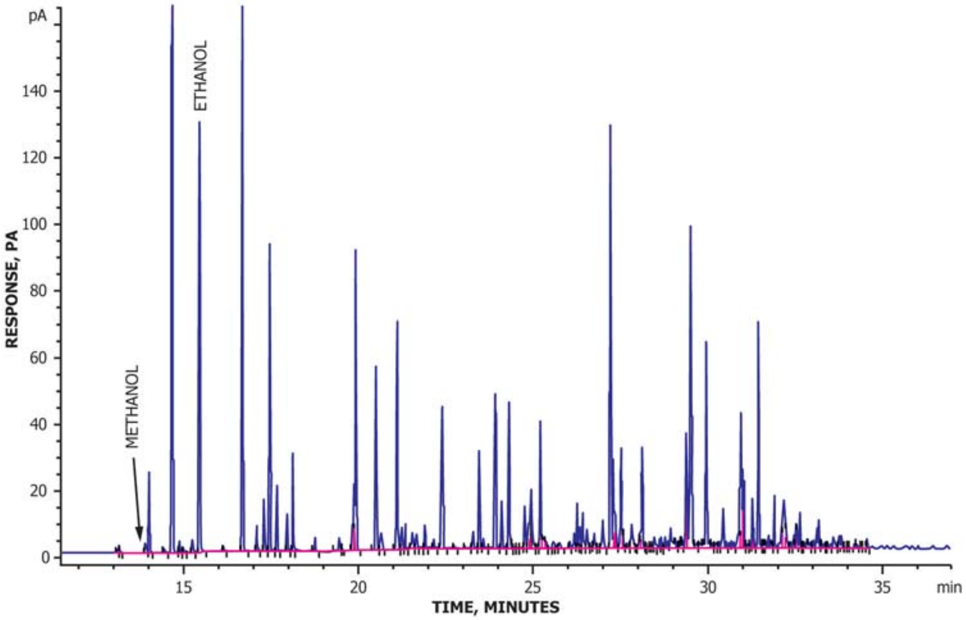


FIG. 6 Sample Chromatogram of a 20 % Ethanol Blend (E20) on 150 m Column

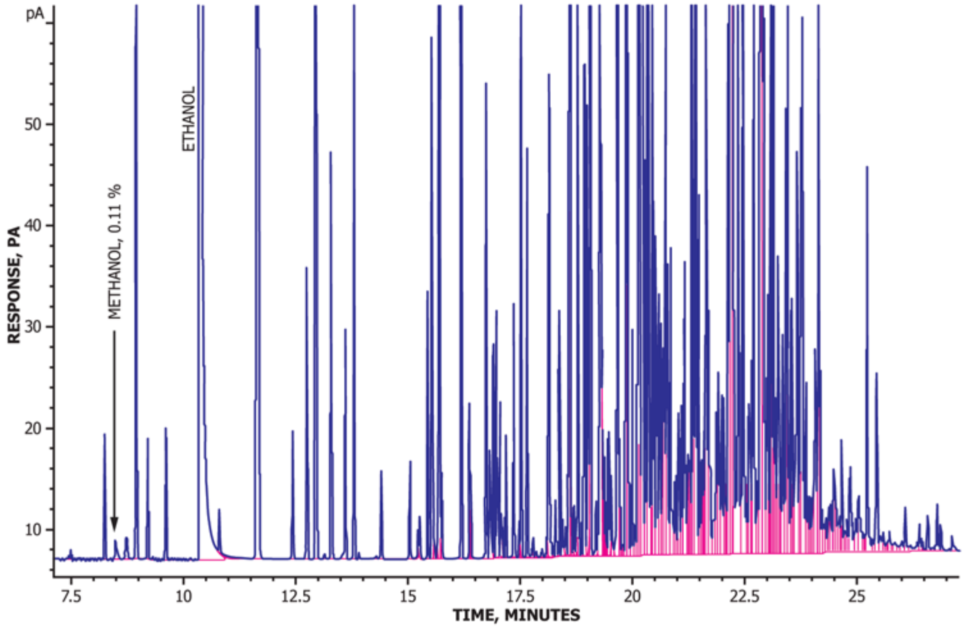


FIG. 7 Sample Chromatogram (Expanded View) of 20 % Ethanol Blend on 100 m Column

13.5 For the volumetric concentration of the alcohol, calculate as follows:

$$V_i = \frac{M_i \times D_s}{D_i} \tag{8}$$

where:

V_i = % by volume of component i,
M_i = % by mass of component i from Eq 7,
D_i = density of component i at test temperature t as found in Table 3, and
D_s = sample density at test temperature t as determined by Test Method D1298 or D4052.

14. Report

14.1 Report the purity of the individual alcohols to the nearest 0.01 % by mass using Eq 7 or nearest 0.01 % by volume using Eq 8 and reference this test method.

15. Precision and Bias[4]

15.1 *Precision*—The precision of this test method as determined by the statistical examination of the interlaboratory gas chromatographic test results of denatured fuel ethanol is as follows:

15.1.1 *Repeatability*—The difference between successive results obtained by the same operator with the same apparatus under constant operating conditions on identical test materials would, in the long run, in the normal and correct operation of the test method exceed the following values only in one case in twenty.

Component	Repeatability[A] Range, % by Mass	Repeatability, % by Mass
Ethanol	20–99.8	$2.1856{*}X^{-0.6}$
Methanol	0.01–0.6	0.02705(X+0.1037)

[A] where *X* is the mass percent.

TABLE 4 Calculated Precision Values for Ethanol and Methanol

	Amount, % by mass	Repeatability	Reproducibility
Ethanol	20	0.36	2.60
	40	0.24	1.72
	51	0.21	1.48
	68	0.17	1.25
	83	0.15	1.11
	97	0.14	1.01
Methanol	0.1	0.01	0.02
	0.2	0.01	0.04
	0.3	0.01	0.05
	0.4	0.01	0.06
	0.5	0.02	0.07
	0.6	0.02	0.09

15.1.2 *Reproducibility*—The difference between two single and independent results obtained by different laboratories on identical test material would, in the long run, exceed the following values only in one case in twenty:

Component	Reproducibility[A] Range, % by Mass	Reproducibility, % by Mass
Ethanol	20–99.8	$15.708{*}X^{-0.6}$
Methanol	0.01–0.6	0.1209(X+0.1037)

[A] where *X* is the mass percent.

NOTE 4—The data in Table 4 shows repeatabilities and reproducibilities for ethanol and several methanol values obtained using the formulas given in 15.1.1 and 15.1.2.

15.1.3 *Bias*—Since there is no accepted reference material suitable for determining the bias for the procedure in these test methods, bias has not been determined.

16. Keywords

16.1 E85; denaturant; denatured; ethanol; ethanol concentration; fuel ethanol; gas chromatography; methanol

APPENDIXES

(Nonmandatory Information)

X1. GRAVIMETRIC PREPARATION OF CALIBRATION STANDARDS

X1.1 Determine the GC purity of the methanol, absolute ethanol, and heptane. Verify there is no heptane in the other hydrocarbon diluent, and no methanol in the ethanol. Determine mass fraction (X1) water in the ethanol and methanol by one of the approved Karl Fisher test methods. Correct the ethanol and methanol purities according to equation X2.

$$\text{Mass fraction} = \text{mass percent}/100 \qquad (X1.1)$$

$$\text{Corrected mass fraction} = \text{GC purity mass fraction} * (1 - \text{water mass fraction})/1 \qquad (X1.2)$$

X1.2 Chill each component before preparing the standard.

X1.3 Choose a glass vial or bottle that is appropriately sized for the amount of standard prepared. The container should have a narrow mouth and minimal headspace remaining after standard preparation. The cap should have a conical polypropylene liner or a PTFE liner. Also, the container and contents should not exceed the capacity of the analytical balance.

X1.4 Place the bottle and cap on the balance. Equilibrate and tare. Quickly add the least volatile component and cap. Record the mass to the nearest 0.1 mg etc. Repeat for next least volatile component until all have been weighed. Cap quickly.

X1.5 Correct the masses of methanol, ethanol, and heptane for impurities and water. Table X1.1 shows example calculations for an E75.

X1.6 Calculate the mass percent of each component according to Eq X1.3.

$$\text{Mass\%}_{(i)} = \text{Corrected Mass}_{(i)}/\text{Total Mass} * 100 \qquad (X1.3)$$

---

[4] Supporting data (results of the 2011 ILS) have been filed at ASTM International Headquarters and may be obtained by requesting Research Report RR:D02-1749. Contact ASTM Customer Service at service@astm.org.

TABLE X1.1 Example Calculation for 75 % Ethanol Calibration Standard Preparation

	A	B	C	D	E
1	Component	Mass Weighed, g	Mass, g Corrected for Purity	Column C Calculations	Mass Percent (purity and water corrected)
2	Methanol	0.251	0.250	= B2 * C13	0.42
3	Ethanol	44.887	44.67	= B3 * B13	74.83
4	Heptane	5.998	5.97	= B4 * D13	10.00
5	Hydrocarbon Diluent	8.565	8.57	= B5 * E 13	14.35
6	Impurities	NA	0.14	= B2 * C14+ B3 * B14 + B4 * D14 + B5 * E14	0.23
7	Water	NA	0.11	= B3 * B12 + B2 * C12 + B4 * D12 + B5 * E12	0.18
8	Total	59.701	59.701		100.00
9					
10		EtOH purity	MeOH Purity	nC7 purity	Hydrocarbon Diluent
11	GC Purity	0.9976	0.9999	0.9950	1.0000
12	KF Water	0.0024	0.0039	0.0001	0.0000
13	Corrected for water	0.9952	0.9960	0.9949	1.0000
14	Impurities (= 1 - Corrected Purity - KF Water)	0.0024	0.0001	0.0050	0.0000

Corrected Mass$_{(i)}$ = purity corrected weight in grams of a component, and
Total Mass = total amount weighed in grams.

The contribution of the impurities has to be included for the total mass of column C to equal the amount weighed in column B.

X1.7 Shake the vial to thoroughly mix the standard. Store in the refrigerator until time of analysis. (Transfer aliquots to smaller vials with minimal headspace for extended storage.)

X1.8 Analyze and calculate the standards as described in Sections 10 through 13.

X2. GRAVIMETRIC PREPARATION OF A QC SAMPLE

X2.1 Follow the procedure and calculation steps given in Appendix X1, except use gasoline or natural gasoline instead of heptane and other hydrocarbons. An example of an E50 QC sample is provided in Table X2.1.

X2.2 The result directly from the chromatographic analysis is not adjusted for water. To estimate the expected analysis result prior to water correction, normalize the mass percent of each component to the total before water, per Eq X2.1.

$$\text{Mass before water correction} = \text{mass\%}_{(i)}/(100 - \%\ \text{water})*100 \tag{X2.1}$$

TABLE X2.1 Example Calculation for E50 Check Standard Preparation

	A	B	C	D	E	F
1	Component	Mass Weighed, g	Mass, g Corrected for Purity	Column C Calculations	Mass Percent (corrected)	Expected Result from GC before Water Correction
2	Methanol	0.200	0.200	= B2 * C12	0.31	0.31
3	Ethanol	32.812	32.68	= B3 * B12	50.65	50.70
4	Gasoline	31.507	31.49	= B4 * D12	48.81	48.86
5	Impurities	NA	0.08	= B2 * C13 + B3 * B13 + B4 * D13	0.12	0.12
6	Water	NA	0.07	= B3 * B11 + B2 * C11 + B4 * D11	0.11	NA
7	Total	64.519	64.519		100.00	100.00
8						
9		EtOH purity	MeOH Purity	Gasoline		
10	GC Purity	0.9976	0.9999	1.0000		
11	KF Water	0.0017	0.0002	0.0005		
12	Corrected for water	0.9959	0.9997	0.9995		
13	Impurities (= 1 –Corrected Purity – KF Water)	0.0024	0.0001	0.0000		

X3. FACTORS THAT AFFECT PRECISION AND ACCURACY

X3.1 *Sample Integrity*—A representative sample is needed to accurately quantify the source material. Sample integrity shall be maintained from the source to the lab. Once in the lab, samples should be chilled prior to sub-sampling. Samples and standards intended for extended use should be refrigerated in a sealed vial with minimal headspace.

X3.2 *Injection Technique*—is critical to both precision and accuracy.

X3.2.1 Automatic injection is needed to achieve the best precision. Small injection volumes (0.1 µL to 0.2 µL) and adequate split prevent overload. (Depending on the default chromatograph configuration, use of modified pressure gauges, flow restrictors, or split vent filters may be necessary to achieve the desired split.) Overloading leads to poor resolution and inaccurate analysis.

X3.2.2 Injector leaks, typically at the septum or other injector seal, will cause sample distortion, tailing, and baseline bleed.

X3.2.3 A poorly maintained syringe (such as one that is "sticky" or no longer gas tight) will also give inconsistent injection. Conduct multiple pre-injection syringe washes with the sample and post-injection syringe washes with solvent. As a precaution, do not use a solvent that is also the analyte (that is, methanol, ethanol, or heptane).

X3.2.4 Autosampler solvent and waste bottles should be kept clean. Waste vial inserts/septa can collect residue that deposits on the syringe and contaminates the sample.

X3.3 *Baseline Stability*—An unstable baseline can result from injector/detector leaks, column bleed, and poor carrier gas quality. Routine maintenance of septa, liner, and syringe is recommended to reduce leaks and prevent carryover. To prevent column bleed, a properly conditioned column is needed. Water injected onto the column can also lead to column bleed. Keep the system dry by performing routine blank analyses or bake-out runs. Do not exceed the maximum temperature rating of the column. To maintain carrier gas quality, supplemental gas filters that remove moisture, oxygen, and hydrocarbons are recommended.

X3.4 *Detector Performance*:

X3.4.1 Column placement in the detector is critical to peak shape and detection of heavy compounds.

X3.4.2 Changes in gas quality and pressure fluctuations can lead to variability in detector response. Detector gas filters/purifiers and a stable gas delivery system are needed for optimal performance.

X3.5 *Integration Technique*—Because this Test Method D5501 is a normalized analysis, all peaks shall be integrated to give an accurate result. Integration parameters shall be adequate to pick up even small peaks eluting at the end of the chromatogram. Ethanol shall be integrated across the entire length of the tail. If the integration cuts off the tail, the proportion of the peak that is integrated will vary with concentration. Use tangential skimming to integrate any rider peaks. The only way to insure that a chromatogram is properly integrated is through visual inspection.

X3.6 *Calculations*—Shall be correct in order to accurately quantify the ethanol and methanol content. The most common calculation errors are completed outside of the chromatography software, such as purity correction. It is important to insure that all secondary calculations, including those carried out within a laboratory information management database, are conducted according to Section 13.

X3.7 Accurate calibration standards are needed to generate accurate results. Typical errors include failure to compensate for purity and water content, calculation errors, and poor preparation technique.

X3.8 A conscientious analyst is the best tool for insuring an accurate and precise determination.

X4. GUIDELINES FOR QUALITY CONTROL

X4.1 Confirm the performance of the instrument or the test procedure by analyzing a quality control (QC) sample.

X4.2 Prior to monitoring the measurement process, the user of the method needs to determine the average value and control limits of the QC sample (see Practice D6299 and MNL 7[5]).

X4.3 Record the QC results and analyze by control charts or other statistically equivalent techniques to ascertain the statistical control status of the total test process (see Practice D6299 and MNL 7). Any out-of-control data should trigger investigation for root cause(s). The results of this investigation may, but not necessarily, require instrument recalibration.

X4.4 In the absence of explicit requirements given in the test method, the frequency of QC testing is dependent on the criticality of the quality being measured, the demonstrated stability of the testing process, and customer requirements. Generally, a QC sample should be analyzed each testing day with routine samples. The QC frequency should be increased if a large number of samples are routinely analyzed. However, when it is demonstrated that the testing is under statistical control, the QC testing frequency may be reduced (see Practice D6792 for further guidance on reducing QC testing frequency).

X4.5 The QC sample precision should be periodically checked against the ASTM method precision to ensure data quality (see Practice D6792 for further guidance on use of Test Performance Index for this purpose).

X4.6 If the analyte concentration of the QC sample is known, a t-test may be used to assess bias (see Practice D6299). If the population mean is statistically different from the hypothetical value, investigate and correct the discrepancy.

X4.7 It is recommended that, if possible, the type of QC sample that is regularly tested be representative of the sample routinely analyzed. An ample supply of QC sample material should be available for the intended period of use, and must be homogenous and stable under the anticipated storage conditions.

X4.8 The Q-procedure described in Practice D6299 may be used to determine control limits when switching between different batches of standard.

X4.9 Refer to relevant documents (see Practice D6299, Practice D6792, and MNL 7) for further guidance on QC and control charting techniques.

---

[5] MNL7, *Manual on Presentation of Data and Control Chart Analysis: 8th Edition*, ASTM International, 2010.

ASTM International takes no position respecting the validity of any patent rights asserted in connection with any item mentioned in this standard. Users of this standard are expressly advised that determination of the validity of any such patent rights, and the risk of infringement of such rights, are entirely their own responsibility.

This standard is subject to revision at any time by the responsible technical committee and must be reviewed every five years and if not revised, either reapproved or withdrawn. Your comments are invited either for revision of this standard or for additional standards and should be addressed to ASTM International Headquarters. Your comments will receive careful consideration at a meeting of the responsible technical committee, which you may attend. If you feel that your comments have not received a fair hearing you should make your views known to the ASTM Committee on Standards, at the address shown below.

This standard is copyrighted by ASTM International, 100 Barr Harbor Drive, PO Box C700, West Conshohocken, PA 19428-2959, United States. Individual reprints (single or multiple copies) of this standard may be obtained by contacting ASTM at the above address or at 610-832-9585 (phone), 610-832-9555 (fax), or service@astm.org (e-mail); or through the ASTM website (www.astm.org). Permission rights to photocopy the standard may also be secured from the Copyright Clearance Center, 222 Rosewood Drive, Danvers, MA 01923, Tel: (978) 646-2600; http://www.copyright.com/