Process monitoring and analysis
biofuel workflows

Ion Chromatography • Liquid Chromatography
• Gas Chromatography • Near-Infrared Spectroscopy

Thermo
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Biofuel production
the global challenge

Biofuel is defined as a solid, liquid, or gas fuel derived from biological material. This broad-based class of biofuel compounds can be separated into two categories.

Bioalcohol comes from crops such as corn, sugar cane, wheat, sorghum, and cellulosic plants such as corn stover, wood, and grasses. With the exception of sorghum, these crops are not naturally high in sugars. However, the grains are high in starch, and the rest of the plant is rich in cellulose and hemicellulose. Making the cellulose more accessible to hydrolysis and solubilizing hemicelluloses sugars is currently difficult and expensive. The analytical challenge is quantifying the diverse mixture of sugars present in hemicellulose.
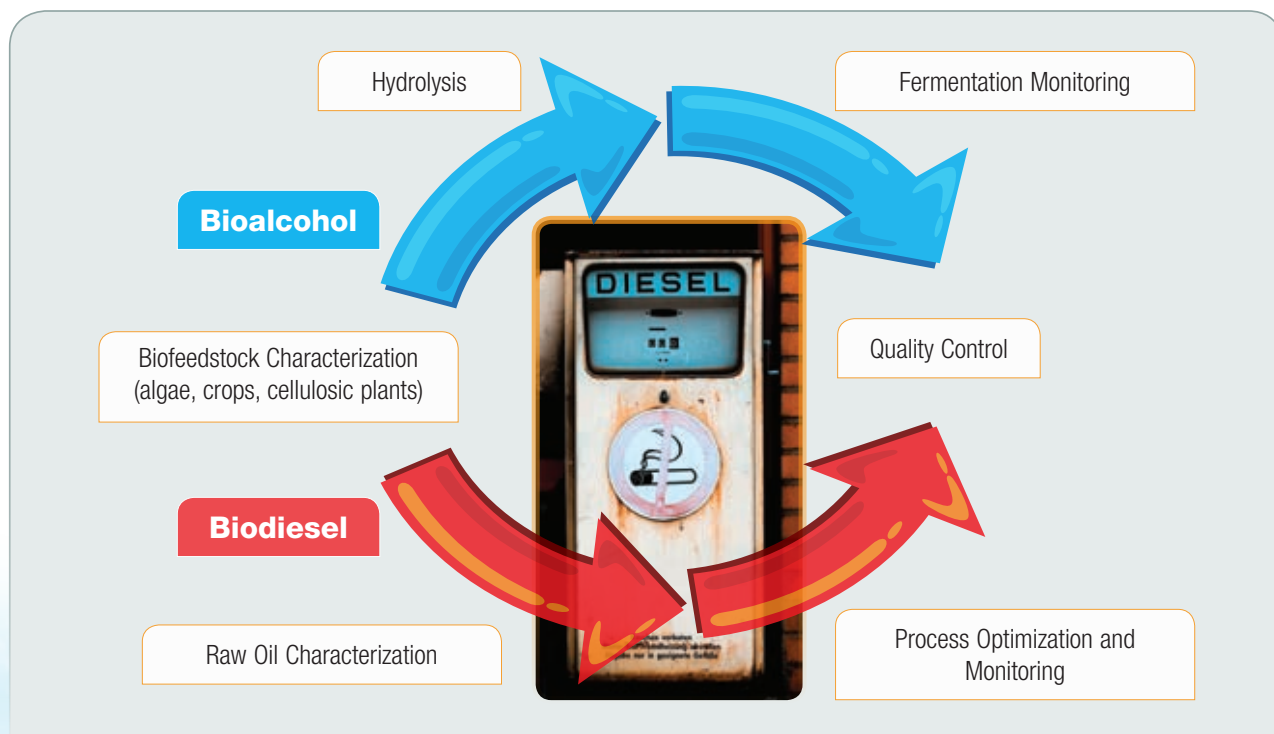
Biodiesel can be produced from plants that contain high amounts of oils, such as soybean, palm, or jatropha. It can also be made using algae. Algae, a single or multicellular plant, can be the source of both sugars for bioalcohol (such as ethanol and butanol) and oils for biodiesel where the need to quantify fatty acid methyl esters (FAMEs) and trace contaminants are key to ensure final product quality.

There are Thermo Scientific™ solutions for every step of your workflow process. Whether the solutions employ near-infrared (NIR) spectroscopy or ion (IC), liquid (LC), or gas chromatography (GC), we can deliver critical information about your biofuel process in a timely manner with a choice of systems and models to best suit your specific application requirements and budget.

Chromatography provides the capability to develop a detailed understanding of the chemical composition and trace contaminant analysis for volatile and nonvolatile samples in every step of the process, while NIR spectroscopy can be used to provide quick answers on feedstock composition on line for real-time process monitoring or final product quality.

Biofuel production
workflows

Analysis Workflow by Application

- Hydrolysis
- Fermentation Monitoring
- Bioalcohol
- Biofeedstock Characterization (algae, crops, cellulosic plants)
- Quality Control
- Biodiesel
- Raw Oil Characterization
- Process Optimization and Monitoring

Analysis Workflow by Product

Sensitivity		Raw Feedstock Characterization	Process Monitoring	Quality Assurance
Lipids	+	Accelerated Solvent Extraction/LC-Charged Aerosol Detection	LC-MS, LC-Charged Aerosol Detection	**FAME** GC-FID
	−		**FAME** NIR	
Carbohydrates	+	Accelerated Solvent Extraction/HPAE-PAD	HPAE-PAD	
	−	NIR, LC-Charged Aerosol Detection	LC-RI, LC-Charged Aerosol Detection, LC-PAD	
Small Molecule	+		**Glycerols** GC HPAE-PAD LC-Charged Aerosol Detection NIR	**Anions, Cations, Group I and II Metals** IC **Methanol** GC-FID NIR
	−			

Systems

System solutions
sample prep and chromatography systems

Sample Preparation
Accelerated Solvent Extraction

The Thermo Scientific Dionex™ ASE™ 150 or 350 Accelerated Solvent Extractor uses elevated temperatures and pressures to rapidly extract water- or oil-soluble components in cellulosic and algal biomass samples.
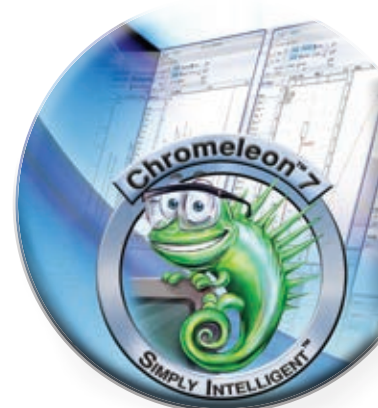
Ion Chromatography

The Thermo Scientific Dionex ICS-5000+ HPIC™ system—with the ability to operate continuously up to 5000 psi—provides fast, high-resolution IC analysis using the latest 4 µm columns.

Liquid Chromatography

The Thermo Scientific Dionex UltiMate™ 3000 LC systems allow you to choose from a wide variety of modules and configurations to create a (U)HPLC instrument configuration that is perfect for your applications.

Gas Chromatography

The Thermo Scientific TRACE™ 1300 Series Gas Chromatograph is the latest technology breakthrough conceived to substantially elevate performance in QA/QC and routine laboratories.

System solutions
analyzers and detectors

NIR Spectroscopy

No matter what the sample, the Thermo Scientific Antaris™ II FT-NIR Analyzer provides robust and reliable data collection for at-line, on-line, and in-line analysis. Analyze raw feedstock by reflection using the internal integrating sphere or liquids with the internal temperature-controlled transmission module. Perform process monitoring with fiber optic probes.

IC or LC: Pulsed Amperometric Detection (PAD)

Electrochemical detection provides high sensitivity detection for analytes that can be reduced or oxidized. In pulsed amperometric mode, sensitive and quantitative results for carbohydrates can be obtained for raw material characterization and process monitoring using established applications.

LC: Charged Aerosol Detection

Charged aerosol detection provides near universal detection independent of chemical structure for nonvolatile and many semivolatile analytes, making it ideal for use with carbohydrates and lipids. Utilized for primary detection or to provide data complementary to UV or MS, this flexible detection method works well for analytical R&D and manufacturing QA/QC.

IC: Conductivity Detection

Designed to measure ionic species in eluents, conductivity detection is especially useful for analytes that lack UV chromophores. When combined with electrolytic suppression, it provides excellent sensitivity and selectivity for numerous ionic species, both organic and inorganic.

GC: Flame Ionization Detection (FID)

Flame ionization detectors are highly efficient and provide a wide linear range and sensitive detection of organic gas and vapor compounds.

Biofeedstock characterization

Accurate, precise compositional analysis of biomass is critical for understanding and assessing biomass conversion technology. Analytical methods that provide a high degree of confidence are required for accurate yield and mass balance calculations, which in turn are necessary for sound cost estimates for biofuel production.


FIGURE 1. The partial least squares (PLS) calibration curve for measurement of xylose demonstrates the capability of NIR spectroscopy to provide rapid analysis of critical components of process feedstock in a few seconds. The primary concentration values for NIR method development are supplied by the chromatographic methods described below.
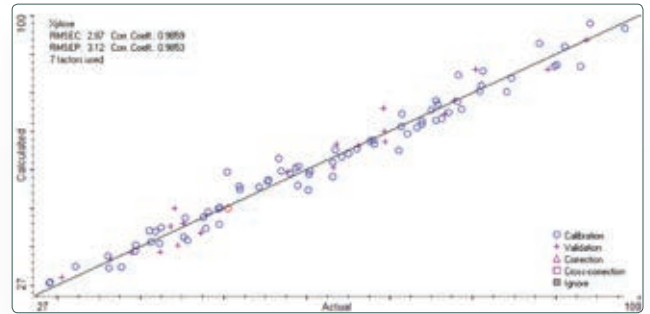
FIGURE 2. Here, hydroxymethylfurfural (HMF), a byproduct and inhibitor of ethanol processing, was detected in acid-hydrolyzed corn stover without interference from the other sugars using high-performance anion-exchange chromatography (HPAE)-PAD. Total run time to provide high sample throughput using a Thermo Scientific Dionex CarboPac™ PA 1 Column was 15 minutes, suggesting this method is ideal for on-line monitoring of HMF during biomass processing.
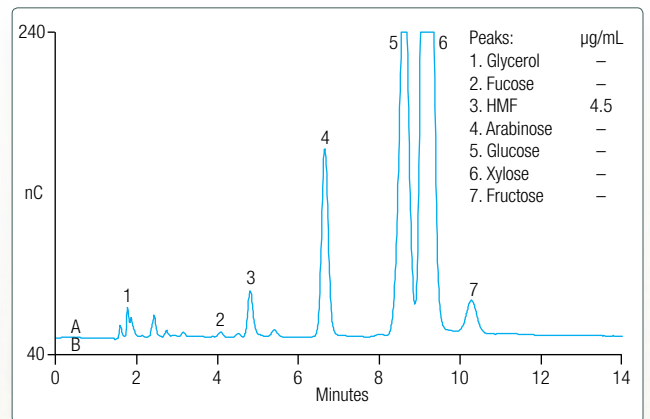

FIGURE 3. This reversed-phase analytical method effectively characterizes lipid samples obtained from algae oil extracts. High-performance LC (HPLC) with charged aerosol detection has the sensitivity to detect low-level compounds for the researcher or analytical chemist, and has reduced chemical requirements (analytes are only required to be nonvolatile) to allow for a broad range of molecular species to be measured.
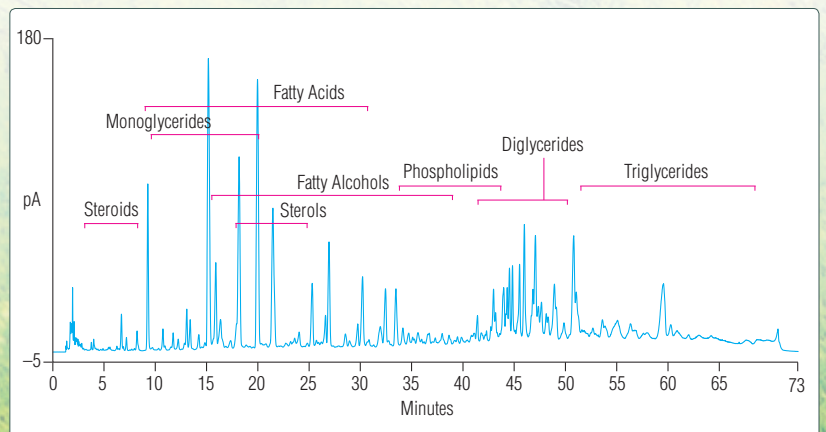
Bioalcohol
fermentation monitoring

A critical step in the development of cellulosic fuels is determining the most favorable conditions for converting complex carbohydrates into fermentable sugars with enzymatic hydrolysis. These reactions typically last up to four days or more, during which time the complex mixtures of carbohydrates, organic acids, and other fermentation inhibitors must be analyzed. Optimization of fermentation processes is critical for maximizing the yields of the final product while ensuring consistent product quality, even during scale-up of biofuel production.
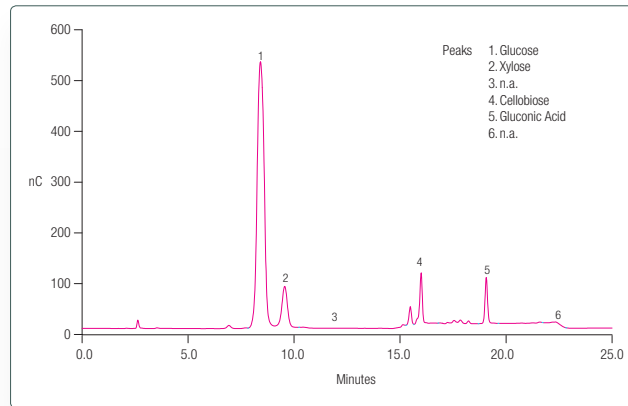
FIGURE 4. Saccharification analysis requires high sensitivity to monitor the enzymatic activity as biomass is hydrolyzed to sugar. Fast run times in which complex mixtures of sugars and byproducts, such as organic acids, are fully resolved is critical for routine, accurate quantitation by HPAE-PAD.
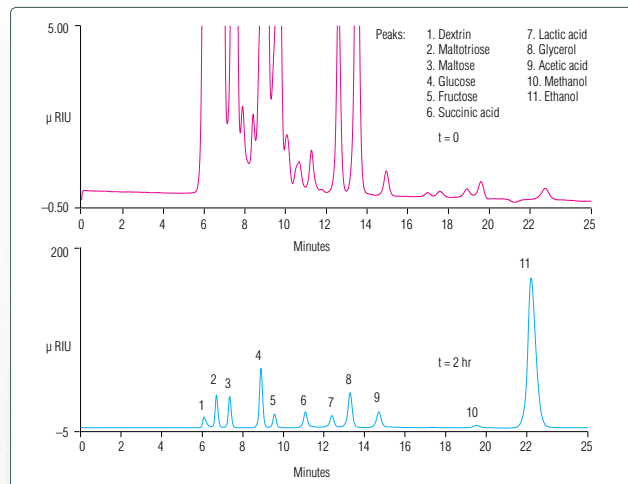
FIGURE 5. During the fermentation process, three key parameters (including eight components) can be easily monitored and quantitatively analyzed by HPLC-RI: 1) the amount of ethanol being produced by HPLC-RI; 2) the amount of fermentable sugars (dextrin, mallotriose, maltose, and glucose) in the fermentation broth; and 3) the concentration of unwanted byproducts (lactic acid, acetic acid, and glycerol) that are produced.

quality control

ASTM International maintains approved written analytical procedures for assuring the quality of denatured fuel alcohols. These regulatory tests are intended to establish minimum quality specifications that all bioalcohol producers must meet to distribute fuel alcohol.
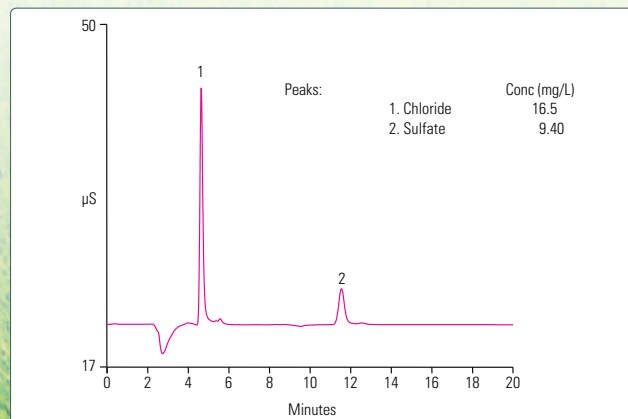
FIGURE 6. The results shown here are from a direct injection IC approach to determine total and potential sulfates and total chloride in butanol. Run time was under 15 minutes using a Thermo Scientific Dionex IonPac™ AS22 Carbonate Eluent Anion-Exchange Column and suppressed conductivity detection.
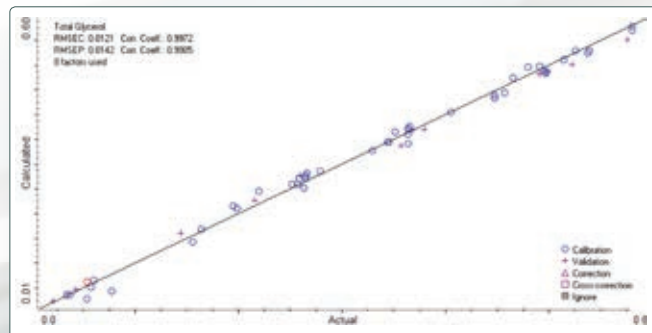
Biodiesel
process monitoring and optimization

Efficient production of biodiesel from microalgae requires analysis of all cell products, including carbohydrates, lipids, and proteins. A complete characterization of the carbohydrate breakdown products is essential for nutrient recycling to determine which sugars are best absorbed by the algae.

FIGURE 7. This separation profile of carbohydrates in microalgae samples shows that more than a dozen peaks were observed. Because many mono- and disaccharides have identical mass-to-charge ratios, IC HPAE-PAD profiles of carbohydrate standards were compared with the sample profile. Comparison of their retention times with monosaccharide standards using a Dionex CarboPac MA1 Column helped to identify the peaks.

Samples:
1. Mannitol
2. Inositol
3. Xylitol
4. D-arabitol
5. L-arabitol
6. Dulcitol
7. Algae sample
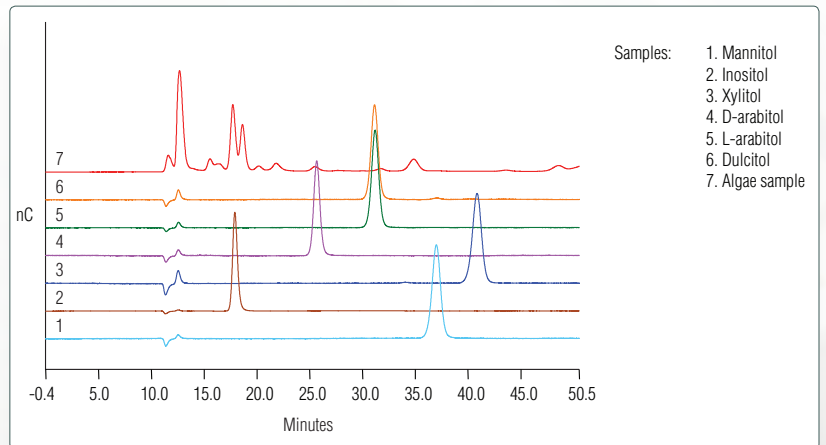
FIGURE 8. Calibration curve for the measurement of glycerol demonstrates the ability of NIR spectroscopy to continuously monitor the biodiesel production process in real time for critical process components.
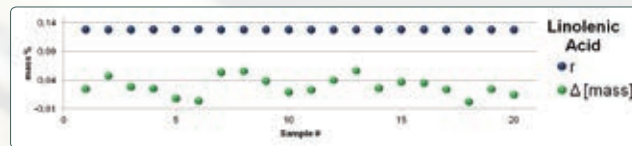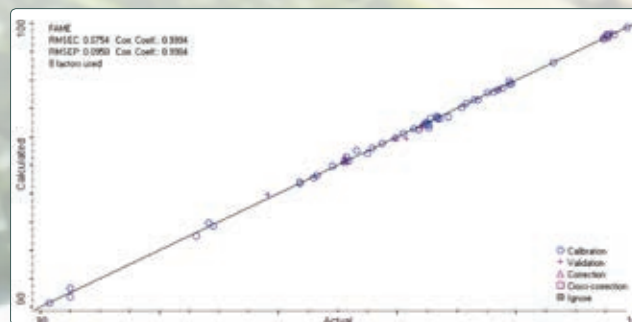
Biodiesel
quality control

A typical process for producing biodiesel is a base-catalyzed transesterificaton reaction of an oil or fat. The oil (triglyceride) is reacted with excess methanol in the presence of sodium hydroxide to yield FAMEs, commonly known as biodiesel. The ability to characterize FAME content and quantify trace contaminants in biodiesel is important for optimizing the biodiesel production process and ensuring final product quality.
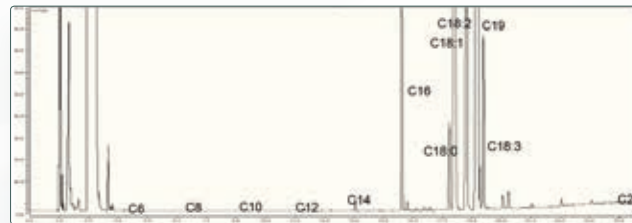
FIGURE 9. This GC chromatogram illustrates determination of FAME and linolenic acid content in a real biodiesel sample, analyzed according to EN 14103 developed by the European Standards Organization (CEN).

FIGURE 10. System repeatability was evaluated on the biodiesel sample; the repeatability of linolenic acid concentration in 20 consecutive injections shows the results well exceed the minimum performance specified in EN 14103, where Δ can be higher than r only in one case in 20 runs. Here r is always higher than Δ.

FIGURE 11. This PLS calibration curve of a FAME analysis shows that final product quality can be determined to a relative error of less than 1% by NIR spectroscopy.

Biodiesel
quality control

Harmful impurities—such as glycerol, methanol, and alkaline earth metals—can lead to damage, clogging, corrosion, poor cold weather performance, and other problematic fuel system conditions. The determination of total glycerol in biodiesel is challenging, as these impurities are not volatile and do not possess chromophores, precluding the use of UV or HPLC fluorescence detection. Left unchecked, high glycerol content may lead to formation of deposits in injector nozzles, pistons, and valves.

Residual methanol in 100% unmodified biodiesel (B100) in even small amounts can reduce the flash point of the biodiesel. Moreover, residual methanol can affect fuel pumps, seals, and elastomers and can result in poor combustion properties.

Alkali and alkaline earth metals in biodiesel may cause corrosion and form soaps, which can cause detrimental deposits in engines and damage engine control systems. To prevent damage from blended fuels, these cations are limited to concentrations less than 5 ppm for sodium and potassium combined, and less than 5 ppm for magnesium and calcium combined, as per ASTM D6751 specifications.


FIGURE 12. A simple, normal-phase method using the UltiMate 3000 HPLC system with the Thermo Scientific Dionex Corona™ ultra RS™ Charged Aerosol Detector provides a fast and accurate measurement of all acylated and free glycerols in a single analysis. In-process biodiesel, finished B100, and mixed petroleum biodiesel (20% biodiesel and 80% petrodiesel, or B20) can be diluted and directly analyzed in under 25 minutes and quantified to the current ASTM D6584 specifications.
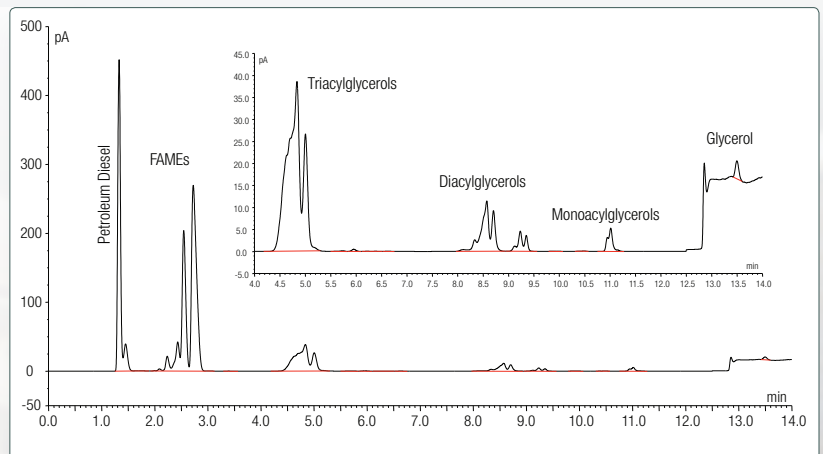

FIGURE 13. IC HPAE-PAD is another well-established method that can determine carbohydrates and glycols without sample derivatization. The Dionex CarboPac MA1 Column provides the selectivity that allows glycerol to be retained longer on this column than on other columns, resulting in the resolution of glycerol from other compounds, and the determination of free and total glycerol in a biodiesel sample. Shown here are results for free glycerol.
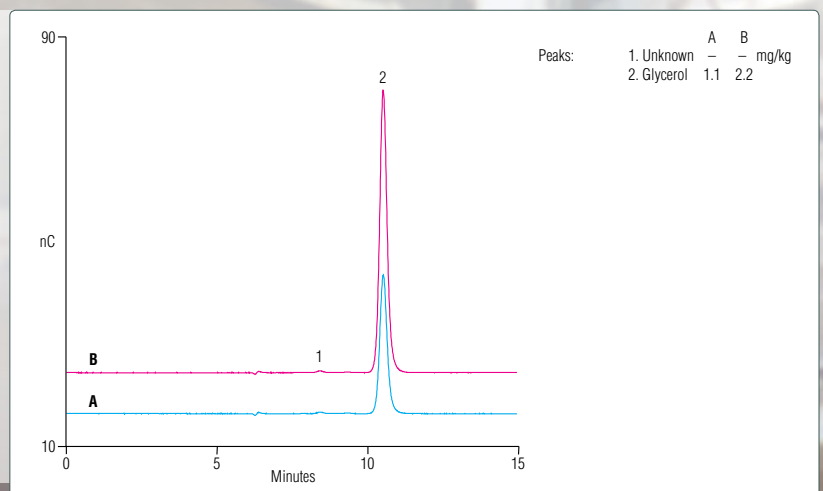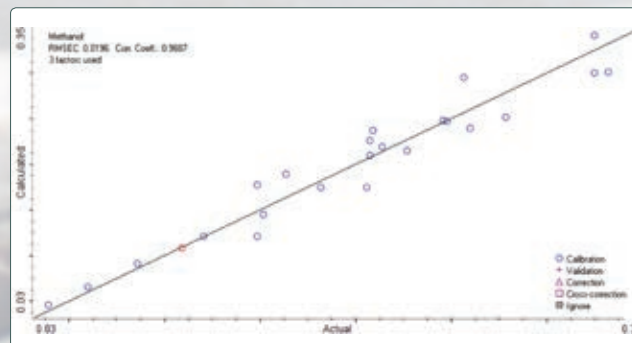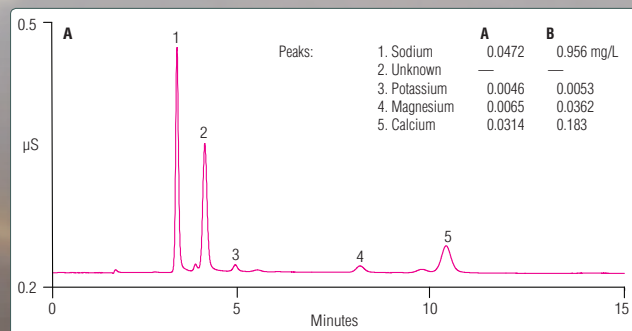
Biodiesel
quality control
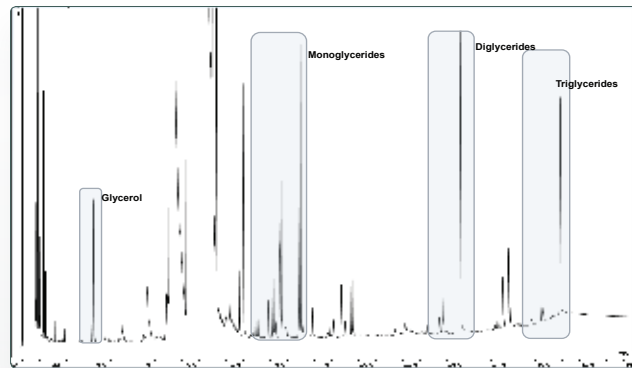
FIGURE 14. This is a chromatogram of a biodiesel sample analyzed with GC/FID according to EN 14105. The areas where glycerol, monoglycerides, diglycerides, and triglycerides were detected are highlighted.
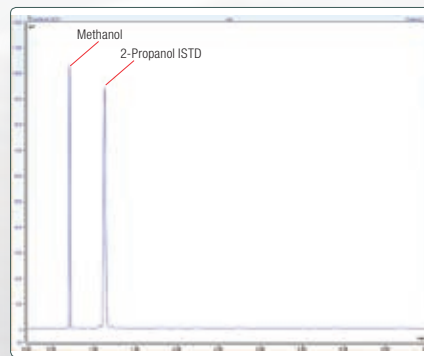
FIGURE 15. Shown is a chromatogram of a biodiesel sample analyzed with GC/FID according to EN 14110 for the determination of methanol content, using 2-propanol as an internal standard.

FIGURE 16. Residual methanol in biodiesel can be measured in a few seconds by NIR spectroscopy below its acceptance limit of 0.2% with an absolute error of 0.02%.

FIGURE 17. In this chromatogram of a B99 (99% biodiesel and 1% petroleum diesel) and B20 extraction, the four cations are well resolved from one another and easily quantified in less than 15 minutes using a Dionex IonPac CS12A Column. The combined sodium and potassium concentration determined in B99 was 0.991 mg/mL with a combined concentration of magnesium and calcium concentration of 0.207 mg/mL, both of which are well below the ASTM limits.

Applications for Process Monitoring and Analysis of Biofuels

Analyte		Application
Biodiesel	AN 40967:	*Analysis of Biodiesel Using the iCAP 6000 Series ICP*
Biomass	AN 363:	*Using Accelerated Solvent Extraction in Alternative Fuel Research*
Carbohydrates	AN 282:	*Rapid and Sensitive Determination of Biofuel Sugars by Ion Chromatography*
	LPN 2827-01:	*Methods for Determining Sugars and Hydroxymethylfurfural in Biomass*
Carbohydrates, Lipids	LPN 2168-01:	*Analysis of Carbohydrates and Lipids in Microalgal Biomass with HPAE-MS and LC/MS*
Chloride, Sulfate	AN 290:	*Determination of Total and Potential Sulfate and Total Chloride in Ethanol According to ASTM Method D 7319*
	AN 297:	*Determination of Total and Potential Sulfate and Total Chloride in Fuel Grade Butanol Per ASTM D7319-09*
FAME	AN 51258:	*Biodiesel (FAME) Analysis by FT-IR*
FAME, Linolenic Acid	AN 10212:	*Determination of Total FAME and Linolenic Acid Methyl Ester in Pure Biodiesel (B100) by GC in Compliance with EN 14103*
FAME, Monoacyl-, Diacyl-, and Triacylglycerols, Glycerol	PN 70046:	*A Single Method for the Direct Determination of Total Glycerols in All Biodiesels Using Liquid Chromatography and Charged Aerosol Detection*
Glycerin	AN 10192:	*Determination of Free and Total Glycerin in B-100 Biodiesel via Method ASTM D6584*
	AN 10215:	*Determination of Free and Total Glycerin in Pure Biodiesel (B100) by GC in Compliance with EN 14105*
Glycerol	AN 255:	*Determination of Free and Total Glycerol in Biodiesel Samples by HPAE-PAD Chromatography*
	AN 1049:	*A Single Method for the Direct Determination of Total Glycerols in All Biodiesels Using Liquid Chromatography*
	AN 51853:	*Determination of Free Glycerol in Biodiesel with the Evolution Array UV-Visible Spectrophotometer*
Group I & II Metals	AN 203:	*Determination of Cations in Biodiesel Using a Reagent-Free Ion Chromatography System with Suppressed Conductivity Detection*
Lipids, Glycerol, Methanol	AN 51544:	*Trace Contaminant Analysis in Biodiesel with an Antaris II FT-NIR Analyzer*
Methanol	AN 10216:	*Determination of Methanol Content in Pure Biodiesel (B100) by Headspace-GC in Compliance with EN 14110*
Oil Content	CAN 301:	*Determination of Oil Content in Biodiesel Feedstock by Accelerated Solvent Extraction*
Sulfur	AN 42164:	*Determination of Sulfur in ULSD, Biodiesel and Jet Fuel using a Thermo Scientific iPRO 5000 Series Analyzer According to ASTM D5453*
	AN PI2044.0207:	*Trace Sulfur Analysis in the Production of Biofuels*

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ISO 9001 REGISTERED

Thermo Scientific Dionex products are designed, developed, and manufactured under an ISO 9001 Quality System.

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