Essential Guide for BIOFUEL ANALYSIS
Tips, Web Tools & Advanced Technologies

• Get Higher Throughput at Lower Cost
• Product Selection Made Easy

www.phenomenex.com/Biofuels
OUR COMMITMENT TO THE BIOFUEL INDUSTRY

Phenomenex is dedicated to supporting the biofuel industry with cutting edge chromatographic tools and resources designed to enhance production, simplify alternative fuels analysis, and lower production costs.

Bioethanol Testing & Quality Assurance
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- GC: Reliably determine free and total glycerin in B100 biodiesel ....................................... p. 8-9

Featured Accessories
- Quickly and easily clean up samples and extend column lifetimes ..................................... p. 10

Column Selection for Biofuels Methods ................................................................. p. 11

Online Resources for Biofuel Analysis
- 1000’s of Applications
- Column Selection Charts
- Technical Notes
- Knowledgeable Technical Consultants

If Phenomenex products in this brochure do not provide at least an equivalent separation as compared to other products of the same phase and dimensions, return the product with comparative data within 45 days for a FULL REFUND.
### Dependably Monitor Ethanol Fermentation Reaction Components

- Easily quantitate ethanol fermentation broth components
- Monitor starches, sugars, organic acids, and ethanol in one run
- Reliably monitor lactic and acetic acids

To maximize ethanol conversion, bioethanol labs monitor key reaction components throughout the fermentation process. With results easily obtained through an isocratic run, Rezex™ ROA-Organic Acid columns separate simple and complex sugars, organic acids, and ethanol found in a fermentation broth sample.

<table>
<thead>
<tr>
<th>Part No.</th>
<th>Description</th>
<th>Dimensions</th>
<th>Unit</th>
<th>Price</th>
</tr>
</thead>
<tbody>
<tr>
<td>00H-0138-K0</td>
<td>Rezex ROA-Organic Acid H+ (8%) HPLC Column</td>
<td>300 x 7.8 mm</td>
<td>ea</td>
<td></td>
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<tr>
<td>03B-0138-K0</td>
<td>Rezex ROA-Organic Acid H+ (8%) HPLC Guard Column</td>
<td>50 x 7.8 mm</td>
<td>ea</td>
<td></td>
</tr>
<tr>
<td>KJ0-4282</td>
<td>SecurityGuard™ Cartridge Holder Kit</td>
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<td>ea</td>
<td></td>
</tr>
<tr>
<td>AJ0-4490</td>
<td>SecurityGuard Carbo-H Cartridges</td>
<td>4 x 3.0 mm ID</td>
<td>10/pk</td>
<td></td>
</tr>
</tbody>
</table>

### Reduce Fermentation Run Times by 50 %

- Increase throughput by reducing run times by 50 %
- Obtain faster results – respond in real-time
- Reduce solvent usage and cost

Rezex ROA achieves excellent baseline separation of typically troublesome Dp3 and Dp4+. This means you can select a shorter column and reduce run times up to 50 %, while maintaining resolution!

#### FAST Fermentation Runs with A Short Rezex Column

<table>
<thead>
<tr>
<th>Part No.</th>
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</tr>
<tr>
<td>AJ0-4490</td>
<td>SecurityGuard Carbo-H Cartridges</td>
<td>4 x 3.0 mm ID</td>
<td>10/pk</td>
<td></td>
</tr>
</tbody>
</table>

Easily quantitate ethanol fermentation broth components
Monitor starches, sugars, organic acids, and ethanol in one run
Reliably monitor lactic and acetic acids

Great Resolution of Dp4+ and Dp3!

Dependably Monitor Ethanol Fermentation Reaction Components

Incorporate Quick and Easy Cleanup!
Filtration of fermentation samples with a 0.20 µm Phenex™ syringe filter can reduce chromatographic interferences due to particulates or cationic impurities. This helps increase run-to-run reproducibility and extend column lifetime!

Learn more on p. 10

#### Incorporate Quick and Easy Cleanup!

Filtration of fermentation samples with a 0.20 µm Phenex™ syringe filter can reduce chromatographic interferences due to particulates or cationic impurities. This helps increase run-to-run reproducibility and extend column lifetime!

Learn more on p. 10

#### Recommended Biofuel Products

<table>
<thead>
<tr>
<th>Biofuel Product</th>
<th>Combinations</th>
<th>Code</th>
<th>Price</th>
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</thead>
<tbody>
<tr>
<td>HPLC Analysis</td>
<td>Rezex ROA-Organic Acid H+ (8%) HPLC Column</td>
<td>300 x 7.8 mm</td>
<td>ea</td>
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<tr>
<td>Column Protection</td>
<td>SecurityGuard Carbo-H Cartridges</td>
<td>4 x 3.0 mm ID</td>
<td>10/pk</td>
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</tbody>
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<td>4 x 3.0 mm ID</td>
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</tr>
</tbody>
</table>
Accurately Measure Ethanol Content in Finished Products

- Completely resolve ethanol and methanol from all denaturant components
- Guaranteed, reliable results

Determination of ethanol and methanol content in final fuel products requires a GC column capable of separating these compounds from all other components (such as fusel alcohols and denaturants). Zebron™ ZB-1 columns provide extremely low bleed levels for highly reproducible, reliable results.

**ASTM Method D5501: Determination of Denatured Bioethanol**

- **Columns:** Zebron ZB-1
- **Part No.:** 7MG-G001-17
- **Influent:** 350 mL/min (Constant Flow)
- **Oven Program:** 45 °C for 7 min to 255 °C @ 30 °C/min (Hold 6 min)
- **Detector:** FID @ 300 °C
- **Sample:** 1. Methanol
  2. Ethanol
  3. Denaturant

**Recommended Biofuel Products**

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</tr>
</thead>
<tbody>
<tr>
<td>7MG-G001-17</td>
<td>Zebron ZB-1 GC Column</td>
<td>100 m x 0.25 mm x 0.50 µm</td>
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</tbody>
</table>

Shorten QA Runs of Bioethanol to 5 Minutes

- Quickly determine ethanol content with shorter run times
- Complete your Certificate of Analysis (COA) in a fraction of the time
- Recommended for methods ASTM D4806, ASTM D5501, and prEN 153767

Standard GC runs of bioethanol with typical columns can take up to 40 minutes per sample to ensure proper separation of methanol and ethanol from denaturant peaks. Zebron ZB-Bioethanol GC columns are designed to streamline this quality assurance (QA) process, with 87% faster run times under 5 min.

**FAST ASTM Method D5501: Reduce Run Times with Short GC Columns**

- **Columns:** Zebron ZB-Bioethanol
- **Part No.:** A) 7EG-G020-22, B) 7HG-G020-22
- **Carrier Gas:**
  A) Hydrogen @ 35 cm/sec (Constant Flow)
  B) Hydrogen @ 25 cm/sec (Constant Flow)
- **Oven Program:**
  A) 45 °C for 2.5 min to 250 °C @ 30 °C/min (Hold 4.5 min)
  B) 55 °C for 1.7 min to 260 °C @ 40 °C/min (Hold 2.67 min)
- **Detector:** FID @ 300 °C

**Recommended Biofuel Products**

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<tbody>
<tr>
<td>7EG-G020-22</td>
<td>Zebron ZB-Bioethanol GC Column</td>
<td>30 m x 0.25 mm x 1.00 µm</td>
<td>ea</td>
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<tr>
<td>7HG-G020-22</td>
<td>Zebron ZB-Bioethanol GC Column</td>
<td>15 m x 0.25 mm x 1.00 µm</td>
<td>ea</td>
<td></td>
</tr>
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</table>
Reliably Determine Free and Total Glycerin in Biodiesel

- Long column lifetimes with temperature stability to 430°C
- Remove contaminants and eliminate carryovers with aggressive temperature programs

To ensure the quality of the biodiesel product, many methods specify high temperature analysis of free and total glycerin. At temperatures above 380°C, however, most fused silica columns become brittle and spontaneously break due to polyimide degradation. Zebron™ ZB-5HT Inferno™ columns are specially designed for thermal stability up to 430°C, ideal for the elevated temperatures needed to resolve and remove high boiling triglycerides in biodiesel.

### ASTM Method D6584: Free and Total Glycerin in B100 Biodiesel

- **Column:** Zebron ZB-5HT Inferno™
- **Dimensions:** 15 m x 0.25 mm x 0.10 µm
- **Part No.:** 7AK-G000-00-GH0
- **Carrier Gas:** Helium @ 1.3 mL/min (constant flow)
- **Detector:** FID @ 380°C
- **Oven Program:** 50 °C for 1 min to 180 °C @ 7 °C/min to 380 °C @ 30 °C/min for 10 min

### Accurately Test B100 Biodiesel

- **Column:** Zebron ZB-XLB-HT Inferno™
- **Dimensions:** 15 m x 0.25 mm x 0.10 µm
- **Part No.:** 7HG-G024-11
- **Carrier Gas:** Helium @ 1.3 mL/min (constant flow)
- **Detector:** FID @ 380°C
- **Oven Program:** 50 °C for 1 min to 180 °C @ 30 °C/min to 380 °C @ 10 °C/min for 10 min

### Recommended Biofuel Products

- **Column:** Zebron ZB-35HT Inferno™
  - **Dimensions:** 15 m x 0.25 mm x 0.10 µm
  - **Part No.:** 7EG-G025-02
  - **Carrier Gas:** Helium @ 3 mL/min (constant flow)
  - **Detector:** FID @ 380°C
  - **Oven Program:** 50 °C for 1 min to 180 °C @ 15 °C/min to 230 °C @ 10 °C/min to 380 °C @ 3 °C/min for 10 min

- **Column:** Zebron ZB-5HT Inferno™
  - **Dimensions:** 15 m x 0.25 mm x 0.10 µm
  - **Part No.:** 7AK-G000-00-GH0
  - **Carrier Gas:** Helium @ 5.79 cm/sec (constant flow)
  - **Detector:** FID @ 380°C
  - **Oven Program:** 50 °C for 1 min to 180 °C @ 15 °C/min to 230 °C @ 10 °C/min to 380 °C @ 3 °C/min for 10 min

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### Bioethanol Fuel Production Source

<table>
<thead>
<tr>
<th>“Sugary” Plants</th>
<th>“Amylaceous” Plants</th>
<th>Cellulosic Material</th>
</tr>
</thead>
<tbody>
<tr>
<td>Beets, Sugarcane</td>
<td>Com, Wheat, Sweet Potato, Sorghum</td>
<td>Grass, Wood</td>
</tr>
</tbody>
</table>

#### Technique
- **HPLC**
  - Fermentation Reaction Monitoring
  - Recommended Column: Rezex™ ROA-Organic Acid HPLC Columns
  - Benefits: Monitor starches, sugars, organic acids, and ethanol all in one run, Ability to reduce run times by 50% with shorter column
- **Ethanol Recovery Testing**
  - Recommended Column: Rezex™ ROA-Organic Acid HPLC Columns
  - Benefits: Quick and reproducible analysis

#### GC
- **Ethanol / Methanol Quality Assurance**
  - Official Methods: ASTM D4806, ASTM D5501, prEN 15376
  - Recommended Column: Zebron™ ZB-Bioethanol GC Columns
  - Benefits: Increase throughput – reduce run times 87%, Resolve ethanol and methanol from denaturant components, Long column lifetimes

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### Biodiesel Fuel Production Source

<table>
<thead>
<tr>
<th>Vegetable Oil</th>
<th>Animal Fats</th>
<th>Recycled Feeds</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soybean Oil, Canola Oil, Corn Oil</td>
<td>White Grease, Poultry, Tallow</td>
<td>Yellow Grease</td>
</tr>
</tbody>
</table>

#### Technique
- **GC**
  - Free and Total Glycerin Determination
  - Recommended Column: Zebron™ ZB-SHT Inferno™
  - Benefits: Temperature stability to 430 °C, Easily remove contaminants and carryovers with aggressive oven ramps, Long column lifetimes

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**Learn more about recommended biofuel accessories at www.phenomenex.com/Biofuels**

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