



# Reliable Analysis of Glycerin in Biodiesel Using a High-Temperature Non-Metal GC Column

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Recently biodiesel has emerged as a leading alternative fuel source because it is easily derived from common feedstocks and can be used in unmodified diesel engines. To support the growth of the biodiesel industry, the United States' American Society for Testing and Materials (ASTM) and the European Deutsches Institut für Normung (DIN) outlined physical and chemical tests to specify the minimum quality standard for biodiesel fuel used in modern diesel engines (1).

Arguably the most critical test for biodiesel is the measure of glycerin content. Glycerin is the major byproduct of the biodiesel production process, called transesterification, where oils and fats are reacted with an alcohol to produce fatty acid methyl esters (FAMES). High glycerin content can lead to a number of fuel problems, such as clogged fuel filters and fuel pressure drops, so its presence must be minimized.

Although GC is the standard analysis technique for this method, it has several inherent challenges. These tests run at very high temperatures and standard fused silica columns are not engineered to withstand temperatures above 380 °C. At temperatures above 380 °C, the polyimide coating of most fused silica columns starts to degrade, eventually becoming brittle and inflexible.

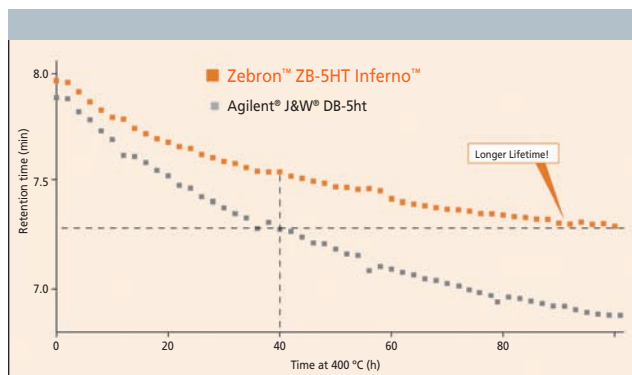
Metal columns, the alternative to fused silica, present other challenges. While metal columns can withstand higher oven temperatures, they are inflexible, difficult to use and require special tubing cutters. In addition, they often develop leaks due to the expansion and contraction that occurs during oven heating cycles and are highly active to acids and bases. Thus, using metal columns might compromise the accuracy of the biodiesel analysis.

Phenomenex, Inc. has recently developed unique fused silica columns designed specifically for high-temperature analysis. These columns, called the Zebtron ZB-1HT and ZB-5HT Inferno, are specially processed to be thermally stable up to 430 °C. Their stationary phases and polyimide coating are more rugged and can withstand higher temperatures than conventional columns. This article compares the lifetime and stability of the Zebtron Inferno column with the leading fused silica columns and presents analysis results on the Zebtron column, using the ASTM D6584 method (2).

## Methods

### Lifetime Comparison

For the high temperature lifetime comparison, three columns were compared: Agilent's (J&W) DB-5ht, Varian's VF-5ht, and Phenomenex's Zebtron ZB-5HT. The columns were held at 400 °C for 2 hours. The oven was lowered to 120 °C then 1.0 µL of pentadecane standard was injected and its retention time was



**Figure 1:** Lifetime comparison of the DB-5ht and ZB-5HT Inferno. The comparison was performed on new, unused DB-5ht and ZB-5HT GC columns. Careful measures were taken to ensure that all conditions were similar for both columns.

measured. This process was repeated 50 times, totaling 100 hours at 400 °C for each column tested.

### Bleed Profile

Bleed (pA) was measured using a flame ionization detector (FID) as the GC oven program increased from 120 °C to 400 °C. The GC oven was held at 120 °C for 3 minutes then increased to 320 °C at 30 °C/minute. A null injection was made at 250 °C.

### Biodiesel Analysis

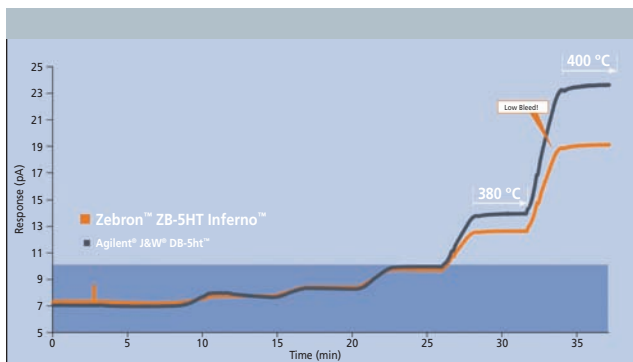
Calibration standards, sample preparation and GC analysis were performed as per ASTM Method D6584.2

## Results and Discussions

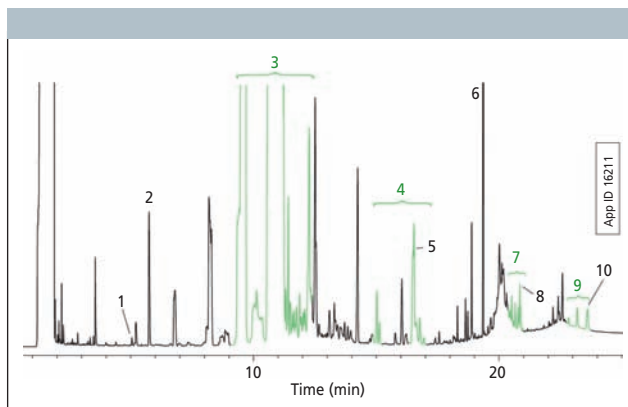
A good indicator of a column's stability is its consistency of hydrocarbon retention time. The pentadecane retention time comparison revealed that the Zebtron ZB-5HT has higher thermal stability than the DB-5ht and VF-5ht. After 40 hours at 400 °C, the VF-5ht column broke (data not shown). In addition, pentadecane has the same retention time on the DB-5ht after 40 hours at 400 °C as on the Zebtron ZB-5HT after 80 hours at 400 °C. This suggests that the ZB-5HT is two times more stable and has twice the column lifetime as the DB-5ht (Figure 1).

Bleed is also an indicator of the stability and lifetime of a GC column. In the bleed comparison test, the DB-5ht showed significantly higher bleed than ZB-5HT at 380 °C (Figure 2). This difference is amplified at 400 °C.

After the initial comparative tests, we conducted the ASTM D6584's free and total glycerin analysis on the Zebtron



**Figure 2:** Bleed comparison of the DB-5ht and ZB-5HT Inferno. The shaded area depicts the bleed criteria for MS certified columns on a MS detector. The ZB-5HT demonstrated lower bleed than the DB-5ht. It meets MS certification limits, even at 360 °C.



**Figure 3:** Reliable analysis of glycerin in biodiesel using a 15m x 0.32mm x 0.10 $\mu$ m Zebtron ZB-5HT Inferno column (Part No. 7EM-G015-02) with a 2.0m x 0.53mm high-temperature guard column. 1  $\mu$ L of the reaction mixture was injected onto a cool-on-column injector connected to a HP 6890 GC (Agilent Technologies) and detected by Flame Ionization (FID). The GC oven program was as follows: 50 °C for 1 min to 180 °C @ 15 °C/min to 230 °C @ 7 °C/min to 380 °C @ 30 °C/min for 10 min. Samples: 1.Glycerol, 2.Butanetriol (ISTD1), 3.Esters, 4.Mono-glycerides, 5.1-Monooleoyl-rac-glycerol, 6.Tricarpin (ISTD2), 7.Diglycerides, 8.1,3-Diolein, 9.Triglycerides, and 10.Triolein.

ZB-5HT column (Figure 3). The calibration curve for each reference components had a correlation coefficient ( $r^2$ ) greater than 0.99 (data not shown).

## Conclusions

To ensure the quality of the biodiesel product, the ASTM Method D6584 specifies a high temperature analysis of free and total glycerin in biodiesel products. However at temperatures above 380 °C, most fused silica columns become brittle and spontaneously break. Our studies suggest that Phenomenex's Zebtron ZB-5HT is well suited for high temperature analyses. When compared to Agilent's (J&W) DB-5ht and Varian's VF-5ht, the Zebtron ZB-5HT Inferno has less bleed, more thermal stability, and longer lifetime.

Having a rugged and durable high temperature GC column will help manufacturers to develop methods that will produce high-quality alternative fuel products. In addition, a column with longer lifetime will help lower the cost per sample of the analysis, thus driving down the cost of production.

## References

- (1) Van Gerpen, J., et al. Biodiesel Analytical Methods, National Renewable Energy Laboratory, 2004.
- (2) ASTM Standard D6584 – 00, "Test Method for Determination of Free and Total Glycerin in B100 Biodiesel Methyl Esters by Gas Chromatography," ASTM International, West Conshohocken, PA, [www.astm.org](http://www.astm.org).

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