

# GC Analysis of Biodiesel for Free and Total Glycerines by ASTM-6584

## Introduction

Rising prices of diesel fuel are creating a market for alternative sources. One of those sources is diesel fuel derived from vegetable oil. Diesel fuel made from plants has the potential to contain glycerols in the form of free glycerin, mono, di and tri glycerides. These compounds have been found to be detrimental to diesel engines so must be removed prior to use. This application note describes a turn-key system for the analysis and automated reporting of free and total glycerines in biodiesel fuel.

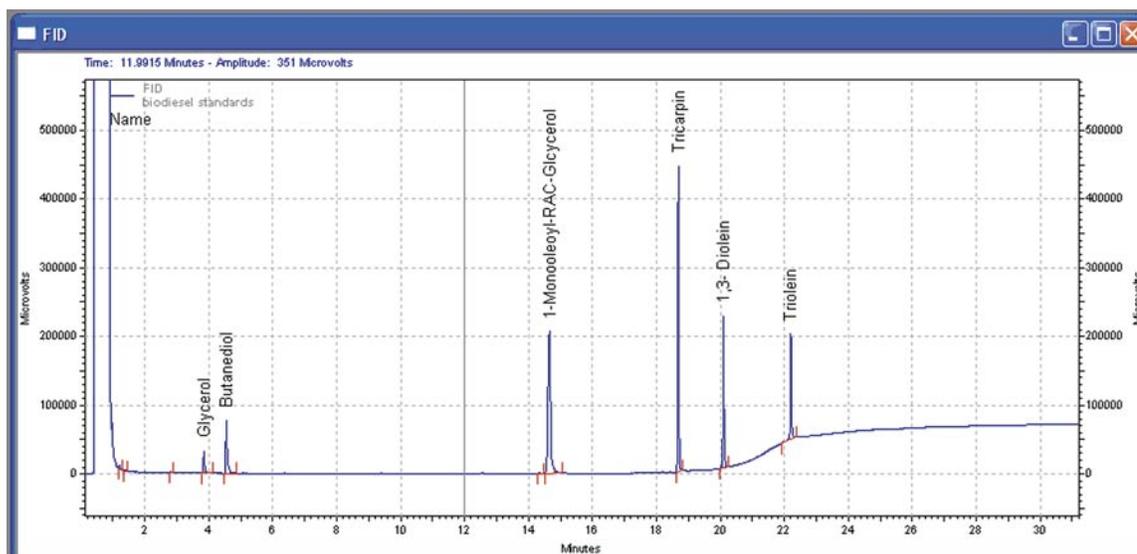


## Application Summary

ASTM-6584 requires a two internal standard, five-point calibration curve be generated for free Glycerine, Monoglycerides, Diglycerides and Triglycerides achieving a linearity of 0.99 r2. Standards are available from Chromatography supply vendors such as Supelco, Belfont, PA. The analytical standards must be derivatized from the free acid form to a methyl ester form so that they will move through a gas chromatographic column prior to analysis. After derivatization, a five-point calibration curve is generated for each of the analyte components of Glycerol, the monoglycerides group, diglycerides group and triglycerides. Unknown biodiesel samples are spiked with an internal standard, derivatized and analyzed. Finally the mono, di and tri group results are summed and reported along with glycerol as free and total glycerin.

## Instrumentation

The instrument configuration adheres to the ASTM method by using the classical temperature programmable On-Column injector and Flame Ionization detector. A 0.32mm id X 12m X 0.1µm 5% capillary column connected to a 2m X 0.53 pre-column will provide the separation. True On-Column injections are made directly into the 0.53 pre-column using a narrow gauge (0.47mm od) needle.



*Biodiesel Standard Chromatogram: 1µl injection of the Supelco Free, mono, di and Triglycerides standard, level 4 concentration.*



## Analytical Conditions

(1ul direct near on-column injection)

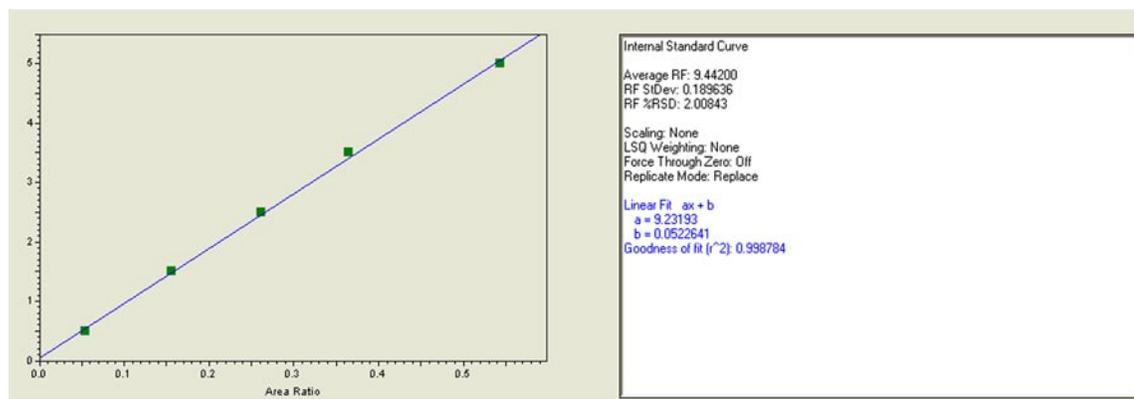
**Injector:** Initial temp: 60°C  
 Rate 1: 15°C/ min. to 190°C  
 Rate 2: 7°C/ min to 230°C  
 Rate 3: 30°C/ min to 380°C, hold 10 min

**Oven:** Initial temp: 50°C  
 Rate 1: 15°C/ min. to 180°C  
 Rate 2: 7°C/ min to 230°C  
 Rate 3: 30°C/ min to 380°C, hold 10 min.

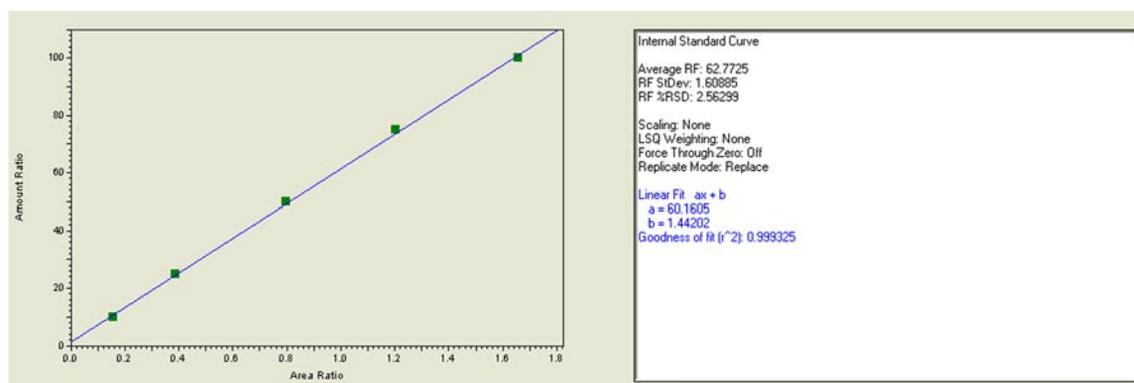
Detector Temp: 380°C  
 Flow: 1.8 ml/min.

## Calibration

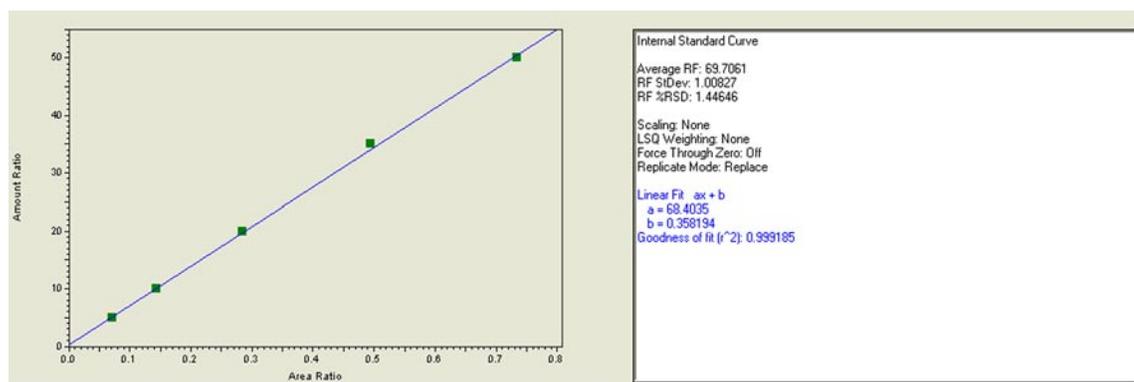
An internal standard calibration of the free, mono, di and tri glycerides was generated using CLASS-VP data analysis software. The calibration curves are presented below.



*Glycerol calibration using 1,2,4-butanetriol as an internal standard*

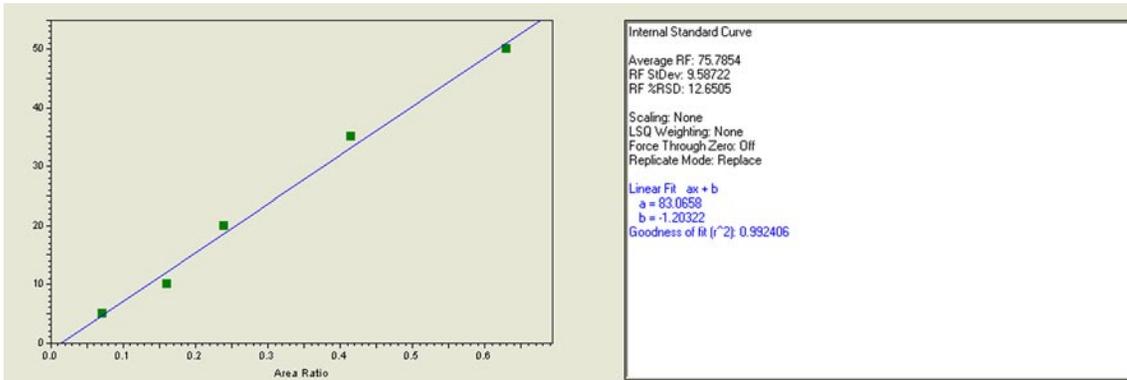


*1-Monooleoyl-RAC-Glycerol calibration curve using Tricarpin as an internal standard*



*1,3-Diolein calibration curve using Tricarpin as an internal standard*

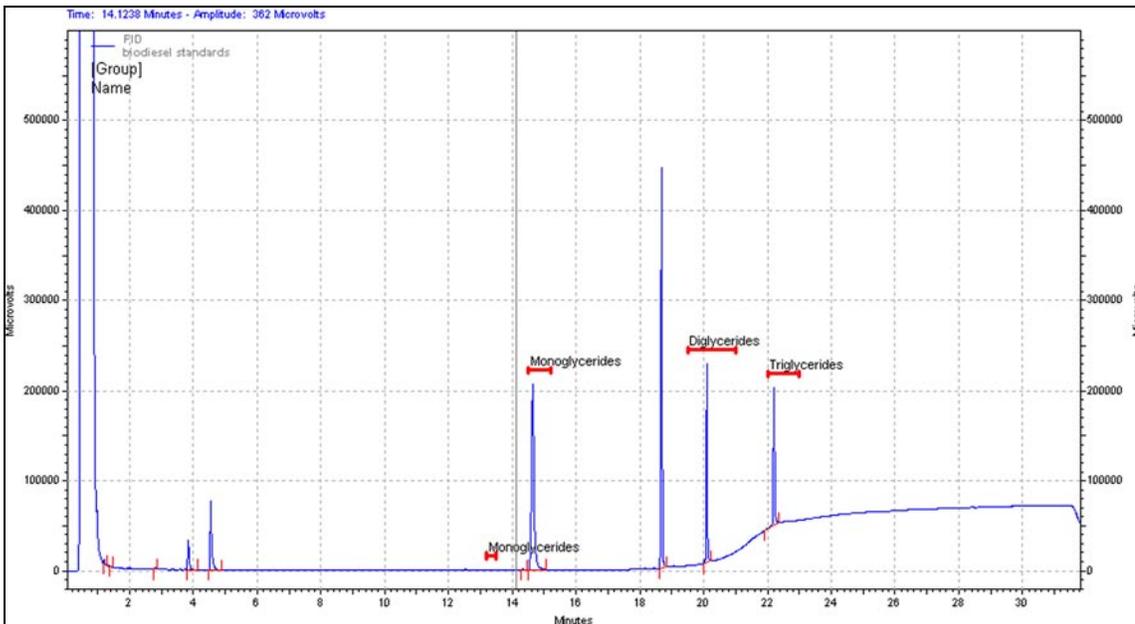




*Triolein calibration curve using Tricarpin as an internal standard*

## Grouping

There are multiple compounds of each of the glyceride classes. To quantitatively account for each of these compounds, the classes are grouped by carbon number. Each group is totaled and the sum treated as a single peak.



*Mono, di and triglyceride groupings*



## Final Report

The method requires that each of the groups be corrected for glycerin content, then summed and reported along with the free glycerin. This is automatically done during batch processing using the Advanced Report function in CLASS-VP. A report will print for each sample. An example report is shown below.

<b>Free and Total Glycerin in Biodiesel Fuel</b>			
File Name:	biodiesel2004-Rep1.dat	Analysis Date:	8/24/2007 10:33:52 ...
Sample ID:	biodiesel standards		
Compound	Raw Conc.	Factor	Final Concentrat...
Glycerol	3.50		3.500
Butanetriol			
1-Monooleoyl-RAC-Glycerol	75.00	0.2591	19.433
Tricarpin			
1,3- Diolein	35.00	0.1488	5.208
Triolein	35.000	0.1044	3.654
		<b>Free Glycerin</b>	3.50
		<b>Free and Total</b>	31.79

## Hardware Package: 220-94683-01

- GC-2010
- Temperature-programmable On-column Injector
- Flame Ionization Detector
- BioDiesel Capillary Column
- EZStart Chromatography Software

ASTM-6584 Standards kit: Includes the derivitizing agent MSTFA, two internal standards and the five-point calibration standards for the free, mono, di and triglycerides

