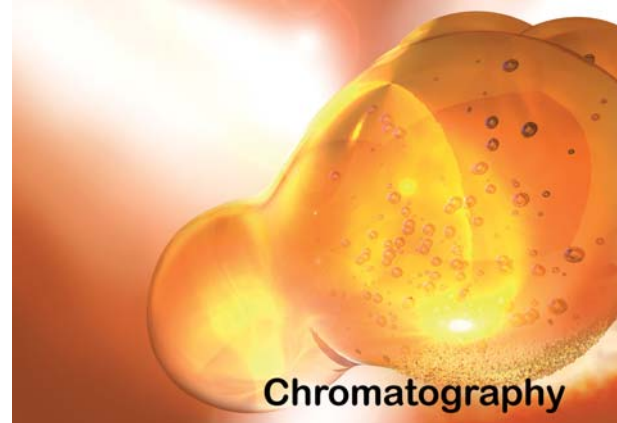


Application Note

Biodiesel Quality Control
According to DIN EN 14105
Determination of free and total glycerol and mono-, di-,
triglyceride contents (reference method)
Part 1: Introduction and sample preparation



Introduction:

Biodiesel is an interesting alternative to the decreasing resources for mineral fuels. It can be manufactured from all kinds of plant oils or even fats from fryers. Because of the high viscosity the direct usage of those regenerative energy resources in diesel cars requires expensive modifications on motor and tank. Therefore the transesterification (Fig. 1) of the glycerol esters to Fatty Acid Methyl Esters (FAME) is preferred. FAMES are less viscous and remain fluid even at low temperatures. They can be used in many diesel cars instead of mineral diesel without changes on the car. Presently the biggest market for biodiesel is the adding to mineral fuels (e.g. B5; up to 5% biodiesel in mineral diesel)

Mainly biodiesel consist of methyl esters of the following fatty acids:

Palmitic acid C₁₆ (saturated)
Stearic acid C₁₈ (saturated)
Oleic acid C₁₈ (unsaturated)
Linoleic acid C₁₈ (poly unsaturated)

The distribution changes dependent on the used plant oil (see table 1). On principal all fatty acids with chain lengths from C₁₄ to C₂₄ might be found (in case of plant oils only the even numbers).

Property Fatty acid in %	Rape seed	Sun flower	Palm tallow	Beef
Palmitic	5	6	42	28
Stearic	1	4	5	19
Oleic	60	28	41	45
Linoleic/Linolenic	30	61	10	5

Tab. 1: Approximate content of fatty acids in different oils and fats.

FAMES are the products of the following reaction:

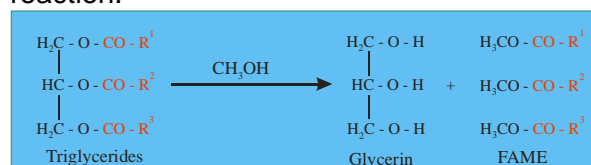


Fig. 1: Transesterification of Tri-glycerols to FAME

Dependent on process parameters the transesterification is more or less quantitative. Generally a certain amount of mono-, di-, and tri-glycerides remains in the biodiesel. Additionally the surplus of methanol and the by-product glycerine must be removed during the biodiesel production. In the quality control according to DIN EN 14105 or ASTM D 6584-00 the contents of the glycerine, mono-, di-, and tri-glycerides are determined. The methanol content is measured with headspace technique described in the DIN EN 14110.

Sample preparation

Gas chromatography is a relative technique. Every component must be calibrated before it can be quantified in unknown samples. In case of biodiesel one mono-, di-, and triglyceride is calibrated representative for the other components of each group.

Table 2 gives an overview of chemicals needed for preparation of standard and unknown samples. Required is a four level calibration of the components 6 to 9. Glycerin (6) is calibrated relative to the internal standard 1 (ISTD1). The glycerides (7-9) are calibrated relative to internal standard 2 (ISTD2).

No	Compound	Alternative naming	CAS No.	Comments
1	MSTFA	N-Methyl-N-(trimethylsilyl)-trifluoroacetamide	24589-78-4	Derivatisation of OH groups
2	Pyridine	Azine	110-86-1	Buffer
3	Heptane		142-82-5	Solvent
4	1,2,4-Butanetriol		3068-00-6	Internal Standard 1
5	1,2,3-Tridecanolyglycerol	Tricaprin	621-71-6	Internal Standard 2
6	Glycerin		56-81-5	
7	1-Monooleoglycerine (Monoolein)	1-Mono [cis-9-octadecenoyl]-rac-glycerol (monoolein)	111-03-5	keep refrigerate -20°C
8	1,3-Dioleoylglycerine (Diglyceride)	1,3-Di[cis-9-octadecenoyl]glycerol (diolein)	2465-32-9	keep refrigerate -20°C
9	1,2,3-Trioleoylglycerine (Triglyceride)	1,2,3-Tri[cis-9-octadecenoyl]glycerol (triolein)	122-32-7	storage below 8°C

Tab. 2: List of components required for biodiesel applications. Possible suppliers are Sigma-Aldrich (Fluka). Due to derivatisation with MSTFA all components must be water free.

Preparation of standard solutions:

Different to DIN EN 14105 we recommend the following standard preparation. Three stock solutions A, B and C are prepared. Later on only the stock solutions A and C are needed for preparation of standard levels and unknown samples.

Stock solution A: Internal Standard

50mg 1,2,4-Butanetriol and 400mg Tricaprin are given into a 50ml flask.
Pyridine is added to 50ml marking.

Stock solution B: Glycerin dilution

50mg Glycerin are given into a 10ml flask.
Pyridine is added to 10ml marking.

Stock solution C:

Olein standard solution with glycerin

200mg Monoolein, 50mg Diolein and 50mg Triolein are given into a 10ml flask.
1ml of the stock solution B is added.
Pyridine is added to 10ml marking.

The three stock solutions can be kept in a refrigerator for weeks. For preparation of the standard levels add the given volumes of stock solution A and C (Tab. 3) into 10ml flasks.

Calibration Level	1	2	3	4
Volume of stock solution A given into a 10ml flask	100µl	100µl	100µl	100µl
Into the same flask add the given volume of stock solution C	10µl	40µl	70µl	100µl

Tab. 3: Preparation of the four standard solutions.

Derivatisation of the calibration mixtures:

Derivatisation of the samples with MSTFA is crucial for improvement of peak shape and chromatographic separation.

100µl MSTFA are added to each of the four flasks prepared according to table 3. Close the flasks, tighten, and shake them well. At room temperature the derivatisation requires minimum 15 minutes. After that time add 8ml of n-heptane to each flask and shake again. The sample is now ready for measurement.

The different levels contain the following concentrations in mass % for the given components:

Calibration Level Conc. in mass %	Glycerin	Mono-olein	Di-olein	Tri-olein
1	0.005	0.2	0.05	0.05
2	0.02	0.8	0.2	0.2
3	0.035	1.4	0.35	0.35
4	0.05	2	0.5	0.5

Tab. 4: Compound concentrations in the four calibration levels (given in mass %)

Preparation and derivatisation of unknown biodiesel samples.

100mg biodiesel is given into a 10ml flask. 100µl of the stock solution A (internal standard) are added to the flask. Then 100µl of MSTFA are added, close the flask, tighten, and shake it well. The derivatisation needs minimum 15 minutes at room temperature. After that time add 8ml of n-heptane to each flask and shake again.

Important:

Derivated standards or samples must be measured within 24 hours.