## **Application Note**



### Analysis of Triglycerides by High Performance Liquid Chromatography with Evaporative Light Scattering Detection

#### Introduction

Triglycerides is one of neutral fat, functioning as energy source, and it is recognized that too much intake may cause arteriosclerosis. Since most of components in Triglycerides have almost no UV absorption, UV detector with short wavelength range or differential refractive index detector is used for Triglycerides analysis. With this method, however, it takes longer time to stabilize the baseline and foreign substances often affect the result. ELSD is known as an effective detection method to solve the problems on fatty analysis including Triglycerides, taking the advantages of its high sensitivity and stable baseline.

This report shows the result of Triglycerides analysis using ELSD.

Keyword: Triglycerides, C18 column, ELSD

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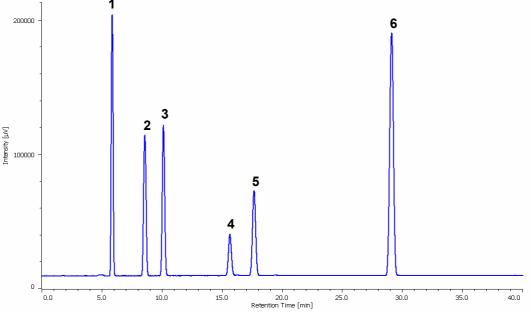
#### Experimental

| 1            |          |                     |  |
|--------------|----------|---------------------|--|
| Equipment    |          | <b>Conditions</b>   |  |
| Pump:        | PU-2089  | Column:             | CrestPak C18S (4.6 mmID x 150 mmL, 5 µm)   |
| Autosampler: | AS-2057  | Eluent:             | A; Acetonitrile, B; THF*   |
| Column oven: | CO-2060  | Gradient condition: | (A/B), 0 min (75/25) $\rightarrow$ 40 min (67/33) $\rightarrow$ 40.05 min (50/50) $\rightarrow$  |
| Detector:    | ELS-2040 |                     | 45 min (50/50) → 45.05 min (75/25) 1 cycle: 60 min   |
|              |          | Flow rate:          | 1.0 mL/min   |
|              |          | Column temp.:       | 40°C   |
|              |          | ELSD condition:     | Nebulizer temp.: 30°C  |
|              |          |                     | Evaporator temp.: 50°C   |
|              |          |                     | Gas flow rate: 1.6 SLM   |
|              |          | Injection volume:   | 10 µL  |
|              |          | Standard sample:    | Trilaurin, Trilinorein, Trimylistin, Triolein, Tripalmitin 1.0 mg/mL each  |
|              |          |                     | Tristearin 0.5 mg/mL   |
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\*) THF solvent does not include any additives.

#### Result

Fig. 1 shows the chromatogram of 6 components of Triglycerides standard mixture. 6 components of Triglycerides were well separated.



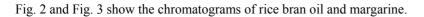
**Fig. 1.** Chromatogram of 6 components of Triglycerides standard mixture 1: Trilaurin, 2: Trilinorein, 3: Trimylistin, 4: Triolein, 5: Tripalmitin, 6: Tristearin

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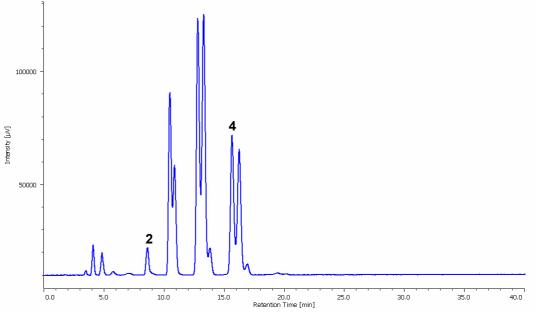
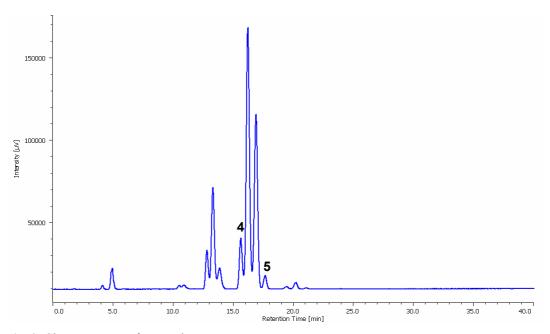
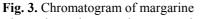


Fig. 2. Chromatogram of rice bran oil The peak numbers are the same as in Fig. 1. Pretreatment: 1.0 g of rice bran oil dissolved in 10 mL aceton was filtrated by 0.45 µm membrane filter.





The peak numbers are the same as in Fig. 1. Pretreatment: 0.5 g of margarine dissolved in 10 mL aceton was filtrated by 0.45 µm membrane filter.