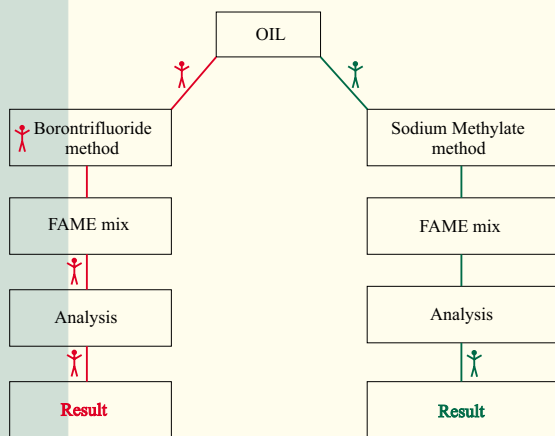


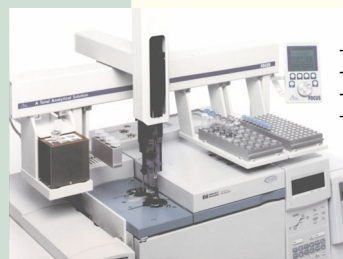
### INTRODUCTION

The preparation of fatty acid methyl esters (FAMES) from fats and oils is one of the most frequently performed chemical reactions in food industry. This reaction is the first step in the analysis of the overall FAME composition of fats and oils. FAME generation is also part of the cis/trans distribution analysis, an analysis that is important due to e.g. health aspects of trans fatty acids. The FAME and cis/trans methyl ester (CTME) analysis requires a manual, time consuming sample preparation step. It is hence clear that there is a significant interest in a faster procedure with reduced manual sample handling by the analyst. The current study aims at developing a faster and automated method for CTME analysis. A fully automated, on-line system for automated methylation using sodium methylate with on-line capillary GC separation is proposed and evaluated.

### MANUAL VS AUTOMATED PROCEDURE



### INSTRUMENTATION



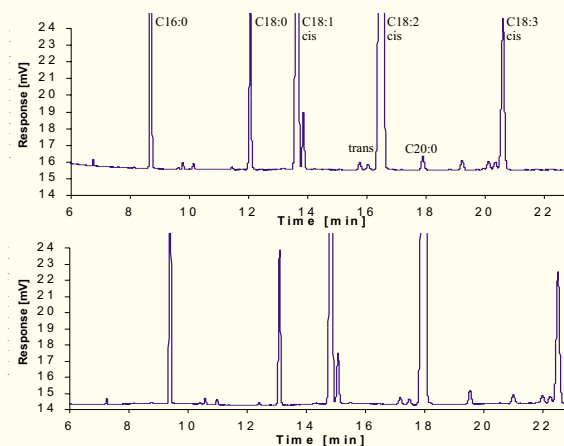
- OPTIC2-200 programmable injector
- FOCUS sample preparation robot
- GC/FID
- CP-SIL88 50m x 0.25mm I.D. d<sub>i</sub>=0.4μm

### TIME GAIN

TABLE 1: Comparison of time spending for sample preparation in CTME analysis.

	Manual BF <sub>3</sub> method	Automated NaOCH <sub>3</sub> method
Time between start of sample preparation and sample ready for injection (min)	35	25
Analyst time required for 15 samples (min)	150	30

### CHROMATOGRAPHIC COMPARISON



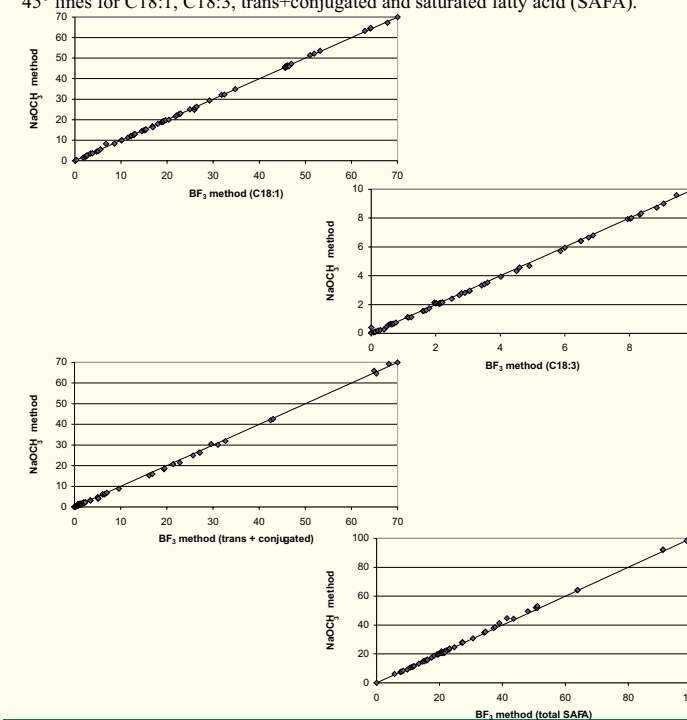
GC-FID chromatograms of the BCR 162 reference sample. Top: New automated on-line analysis with NaOCH<sub>3</sub> methylation. Bottom: Off-line method. FAMES prepared by the BF<sub>3</sub> method.

### TRUENESS

Fatty acid methyl ester	BCR (certified values)		BF <sub>3</sub> n=22		NaOCH <sub>3</sub> n=22	
	%	SD	%	SD	%	SD
C16:0	10.65	0.17	10.61	0.17	10.69	0.117
C18:0	2.87	0.07	2.88	0.07	2.79	0.022
C18:1	24.14	0.28	24.00	0.28	23.94	0.053
C18:2	56.66	0.54	56.06	0.54	56.38	0.068
C18:3	4.68	0.21	4.82	0.21	4.73	0.027

### REAL SAMPLE COMPARISON

In the previous chromatograms the two methods were compared for the BCR 162 reference sample. An additional comparison was also performed for 65 real samples. The analysis reports of these samples contain concentrations of more than 40 (!) compounds. The comparison of the data is shown in a graphical approach where the two methods are plotted against each other. The drawn lines in the graphs give the 45° lines for C18:1, C18:3, trans+conjugated and saturated fatty acid (SAFA).



### CONCLUSION

The proposed automated NaOCH<sub>3</sub> procedure for CTME sample preparation of fat and oil samples is faster than the standard BF<sub>3</sub> method. The manual sample handling time is reduced to approx. 2 minutes per sample. The total sample preparation requires 25 minutes. Because this is fully automated and performed during the analysis of the previous sample, the net sample preparation time is only two minutes. The quantitative results obtained with the two methods are closely similar. Minor differences, typically less than one standard deviation, were seen in a very large set of 65 samples. In analytical terms these differences are not significant. The automated method effects considerable cost-savings because of reduced manual operator time, the increased sample throughput and the reduced use of solvents and reagents. The automated system is robust and user-friendly.