Reproducible FAME Analysis on 100 meter Zebron™ ZB-FAME GC Column

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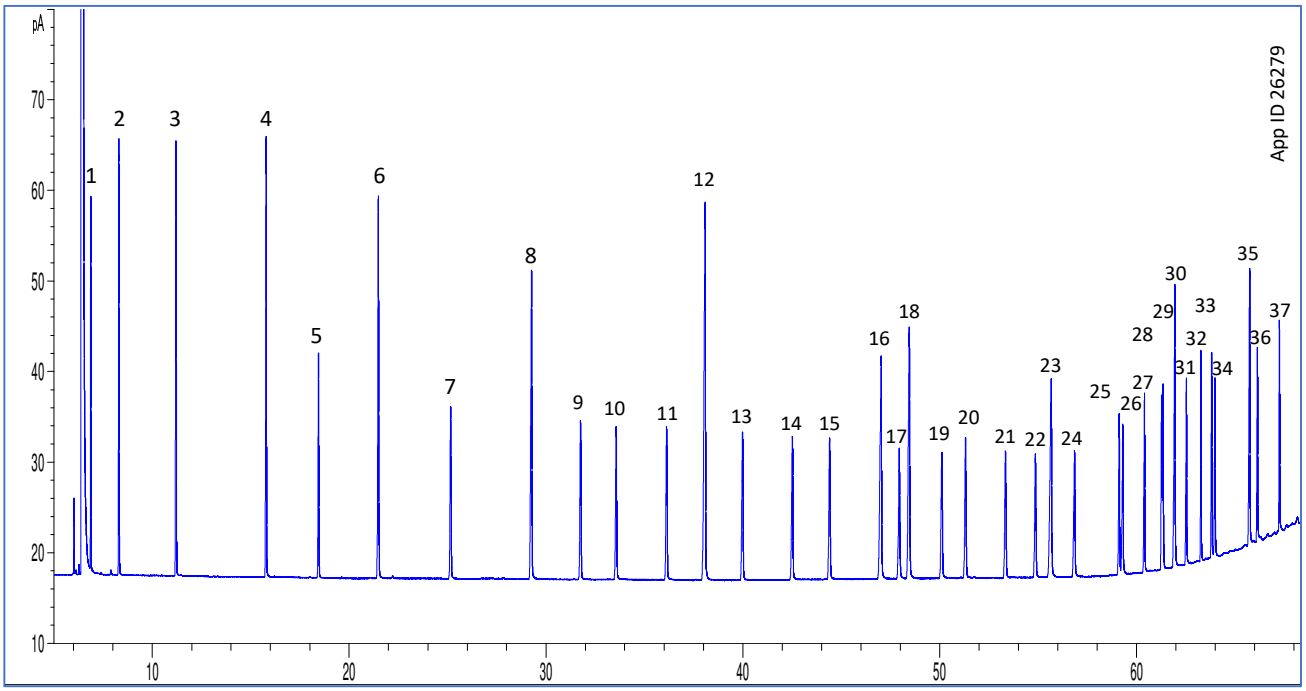
Overview

Fatty Acid Methyl Esters (FAMEs) are derivatized esters of long chain fatty acids and are monitored in food for finger printing authenticity of expensive olive oils, determining trans fat levels, and ascertaining omega-3 and omega-6 content. The separation is extremely challenging because of the presence of cis and trans isomers of the FAMEs that are structurally similar to each other and exhibit similar boiling points. In this study, a highly crosslinked GC stationary phase ZB-FAME was explored for the separation of cis/trans FAME compounds. ZB-FAME GC columns are made of unique, high cyano stationary phase selectivity that can recognize the difference between FAME compounds and isomers that are very similar in physical properties. In addition to providing the required selectivity, ZB-FAME offers a highly crosslinked stationary phase that provides low bleed and high temperature limits of 280 °C . ZB-FAME is available in a traditional 100-meter dimension as well as shorter column dimensions that can provide up to 6 times faster analysis.

GC-FID Conditions

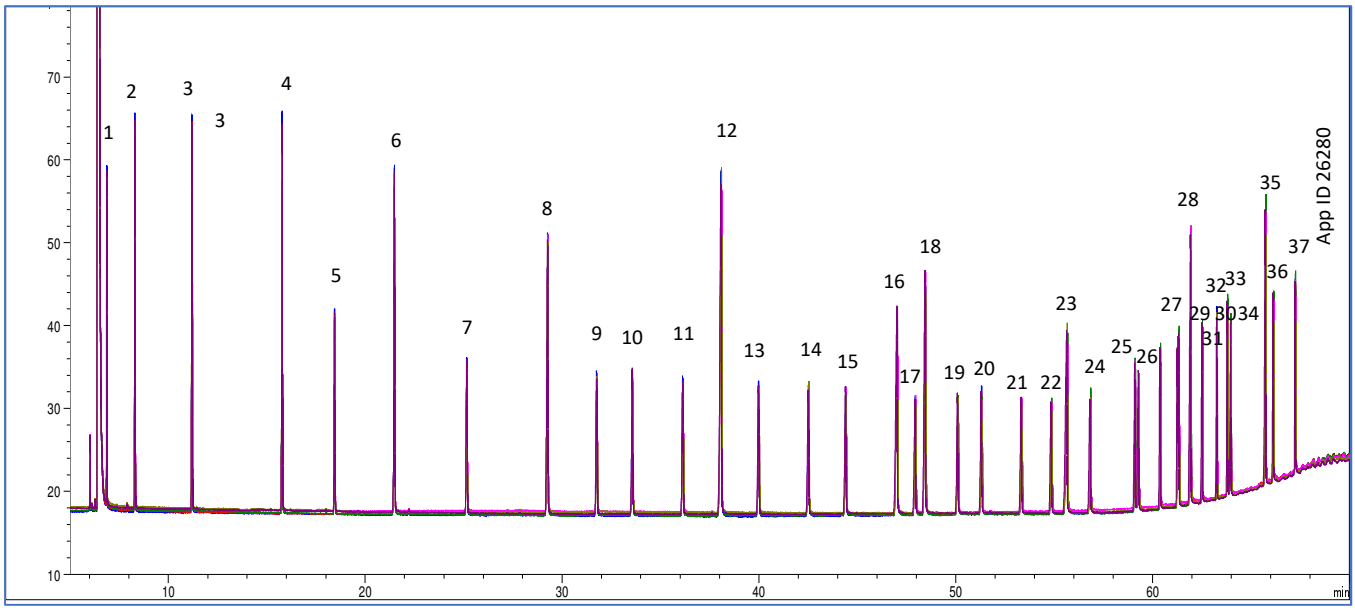
Column (Figure 1 & 2): Zebron ZB-FAME
Dimension: 100 meter x 0.25 mm x 0.20 µm
Part No.: 7MG-G033-10
Column (Figure 3): Agilent® J&W HP-88
Dimension: 100 meter x 0.25 mm x 0.20 µm
Recommended Z-Guard™: 7AG-G000-00-GZK
Injection: Split (50:1) @ 250 °C, 1 µL
Recommended Liner: Zebron PLUS Liner Compatible with Agilent GC Instrument, Single Taper Z-Liner with Wool
Liner Part No.: AG2-0A13-05
Carrier Gas: Hydrogen @ 1.2 mL/min (constant flow)
Oven Program: 100 °C for 5 min, 140 °C @ 3 °C/min for 5 min, 190 °C @ 1.5 °C/min, 260 °C @ 6 °C/min for 5 min
Detector: GC-FID
Temperature: 280 °C

Figure 1: Separation of 37 Component FAME Mix on a 100-meter Zebron ZB-FAME GC Column



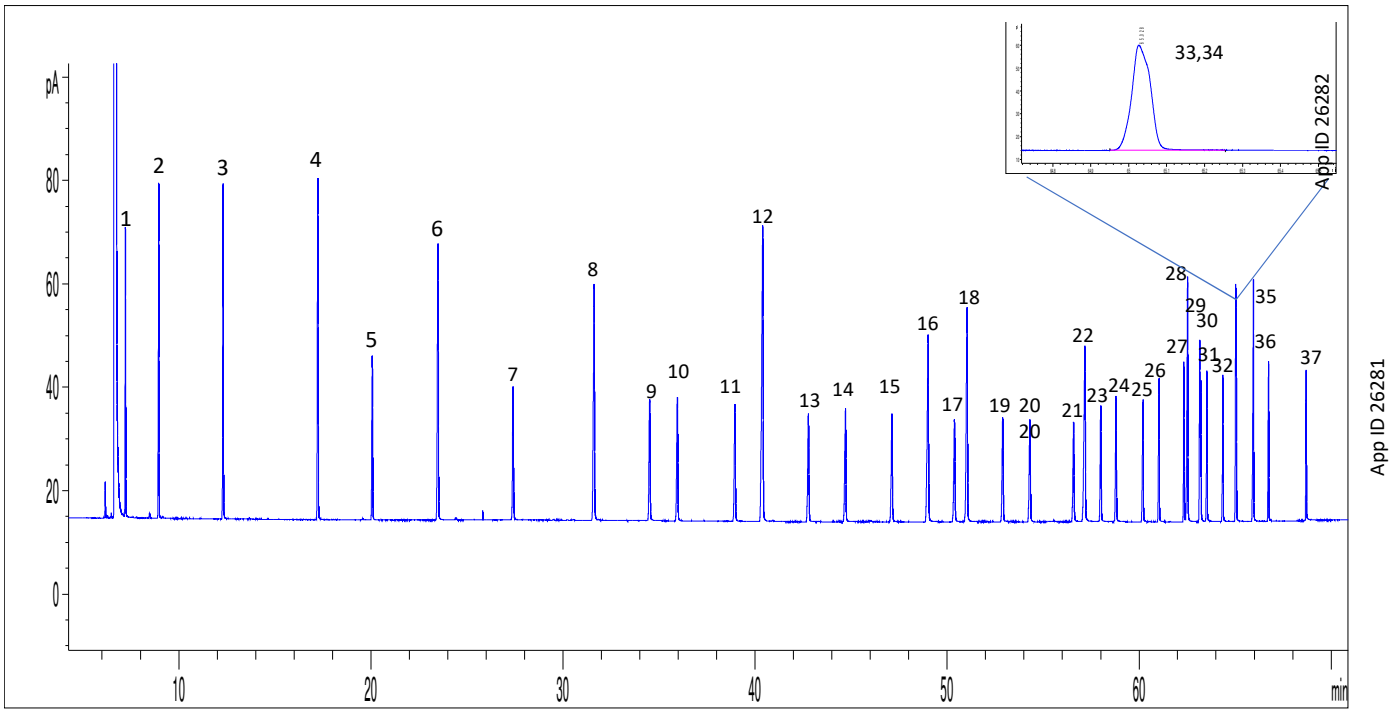
Analyte details in Table 1

Figure 2: Precision of 6 Injections of 37 Component FAME Mix on a 100-meter ZB-FAME GC Column



Same GC Method Parameters as in page 1

Figure 3: Separation of 37 Component FAME Mix on a 100-Meter Agilent® J&W HP-88 GC Column



Same GC Method Parameters as in page 1

Comparative separations may not be representative of all applications.

Table 1.
Peak Area and Retention Time of 37 Component FAME Mix on a 100-meter ZB-FAME GC Column

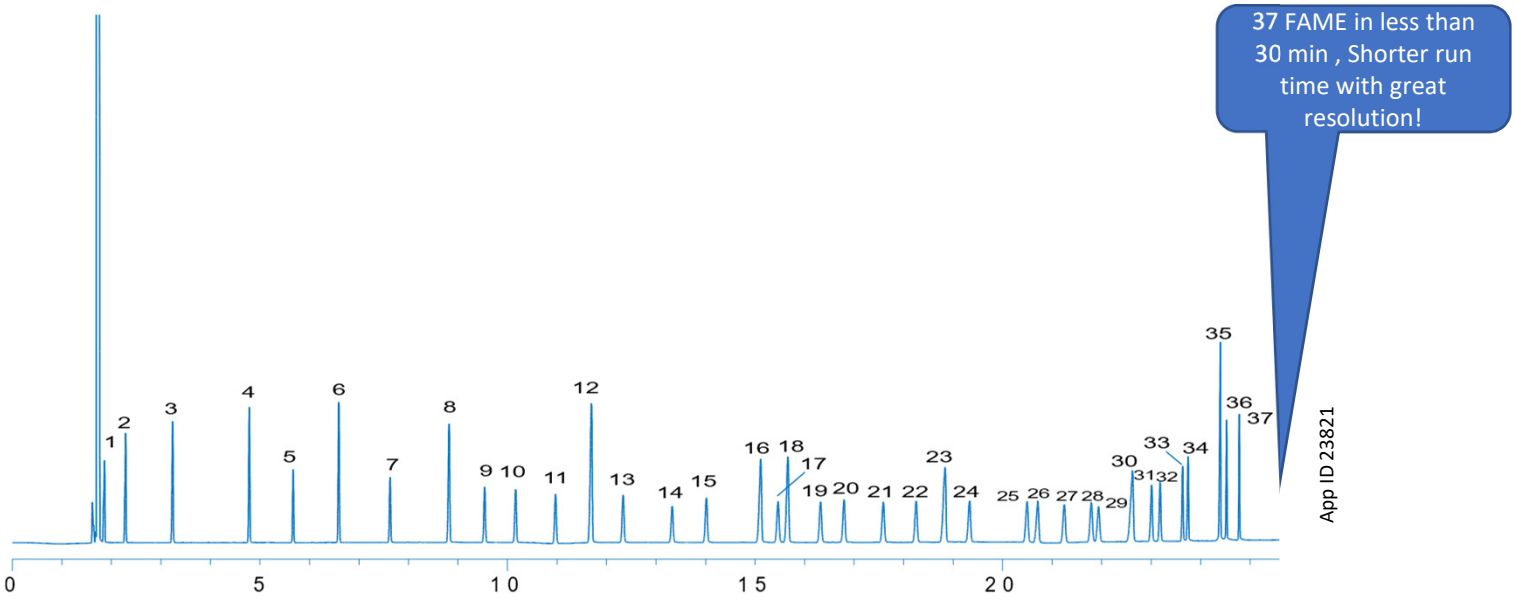
Peak	Analyte Name	Alternate Nomenclature	Resolution	RT (min)	RT %RSD (n=6)	Area %RSD (n=6)
1	Butanoic Acid Methyl Ester	C4:0	11.0	6.87	0.02	1.32
2	Hexanoic Acid Methyl Ester	C6:0	43.5	8.30	0.01	1.51
3	Octanoic Acid Methyl Ester	C8:0	79.9	11.20	0.01	1.96
4	Decanoic Acid Methyl Ester	C10:0	113.1	15.78	0.01	2.38
5	Undecanoic Acid Methyl Ester	C11:0	63.1	18.45	0.01	2.59
6	Dodecanoic Acid Methyl Ester	C12:0	64.4	21.49	0.01	2.72
7	Tridecanoic Acid Methyl Ester	C13:0	67.3	25.16	0.02	2.58
8	Myristic Acid Methyl Ester	C14:0	69.0	29.27	0.02	2.74
9	Myristoleic Acid Methyl Ester	C14:1 cis 9	40.0	31.77	0.03	2.62
10	Pentadecanoic Acid Methyl Ester	C15:0	29.3	33.58	0.03	2.74
11	cis-10-Pentadecenoic Acid Methyl Ester	C15:1 cis 10	40.2	36.14	0.03	2.62
12	Hexadecanoic Acid Methyl Ester	C16:0	27.7	38.09	0.03	2.86
13	Palmitoleic Acid Methyl Ester	C16:1 cis 9	27.2	39.99	0.03	2.80
14	Heptadecanoic Acid Methyl Ester	C17:0	38.6	42.53	0.02	2.86
15	cis-10-Heptadecenoic Acid Methyl Ester	C17:1 cis 10	28.3	44.41	0.02	2.95
16	Stearic Acid Methyl Ester	C18:0	36.5	47.02	0.02	2.95
17	Elaidic Acid Methyl Ester	C18:1 trans 9	12.6	47.95	0.03	2.95
18	Oleic Acid Methyl Ester	C18:1 cis 9	7.2	48.45	0.03	3.05
19	Linolelaidic Acid Methyl Ester	C18:2 trans 9,12	24.1	50.10	0.03	3.30
20	Linoleic Acid Methyl Ester	C18:2 cis 9,12	17.7	51.31	0.03	3.26
21	γ-Linolenic Acid Methyl Ester	C18:3 cis 6,9,12	29.7	53.34	0.03	3.29
22	α-Linolenic Acid Methyl Ester	C18:3 cis 9,12,15	22.8	54.86	0.03	3.24
23	Arachidic Acid Methyl Ester	C20:0	11.0	55.66	0.03	3.11
24	cis-11-Eicosenoic Acid Methyl Ester	C20:1 cis 11	15.9	56.84	0.03	3.08
25	cis-11,14-Eicosadienoic Acid Methyl Ester	C20:2 cis 11,14	37.1	59.11	0.02	3.23
26	Heneicosanoic Acid Methyl Ester	C21:0	3.3	59.29	0.03	3.29
27	cis-8,11,14-Eicosatrienoic Acid Methyl Ester	C20:3 cis 8,11,14	20.4	60.39	0.02	3.15
28	Arachidonic Acid Methyl Ester	C20:4 cis 5,8,11,14	18.3	61.27	0.02	3.19
29	cis-11,14,17-Eicosatrienoic Acid Methyl Ester	C20:3 cis 11,14,17	1.6	61.34	0.02	3.64
30	Behenic Acid Methyl Ester	C22:0	12.0	61.94	0.02	3.39
31	Erucic Acid Methyl Ester	C22:1 cis 13	11.8	62.52	0.02	3.42
32	cis-5,8,11,14,17-Eicosapentaenoic Acid Methyl Ester	C20:5 cis 5,8,11,14,17	17.2	63.27	0.02	3.41
33	cis-13,16-Docosadienoic Acid Methyl Ester	C22:2 cis 13,16	13.3	63.81	0.02	3.46
34	Tricosanoic Acid Methyl Ester	C23:0	4.0	63.98	0.02	3.48
35	Lignoceric Acid Methyl Ester	C24:0	37.1	65.74	0.02	3.79
36	Nervonic Acid Methyl Ester	C24:1 cis 15	8.6	66.13	0.02	3.98
37	cis-4,7,10,13,16,19-Docosahexaenoic Acid Methyl Ester	C22:6 cis 4,7,10,13,16,19	29.7	67.25	0.02	4.28

Summary of Results

This application note demonstrates reproducible retention of FAME components on traditional 100 meter selectivity. **Figure 1** represents complete separation of 37 component FAME mixture on a traditional selectivity after an hour bakeout at 280 °C . **Table 1** presents the precision data for both peak area and retention time on a ZB-FAME 100 meter column. The %RSD for retention time for six replicate injection is less than 0.5% which demonstrates intact stationary phase in ZB-FAME. With conventional high cyano coated FAME analysis columns, this type of reproducibility is not feasible. **Figure 2** represents the overlapped chromatograms of 6 injections with negligible difference in peak area and retention time. On contrary, **Figure 3** shows separation of FAME on an Agilent® J&W HP-88 GC column that lost resolution of peak 33 and 34 after an hour bakeout at 280 °C.

For fast FAME analysis, ZB-FAME is also available in 60, 30, and 20-meter formats. Due to similar phase volume ratio among dimensions, any FAME method can be seamlessly transferred to achieve same resolution, yet a shorter run time. **Figure 4** presents separation of 37 component FAME on a 30 meter column with run time as short as 30 min. Further, with a 20 meter dimension (**Figure 5**), a run time of 11 min was achievable with baseline resolution of all the peaks.

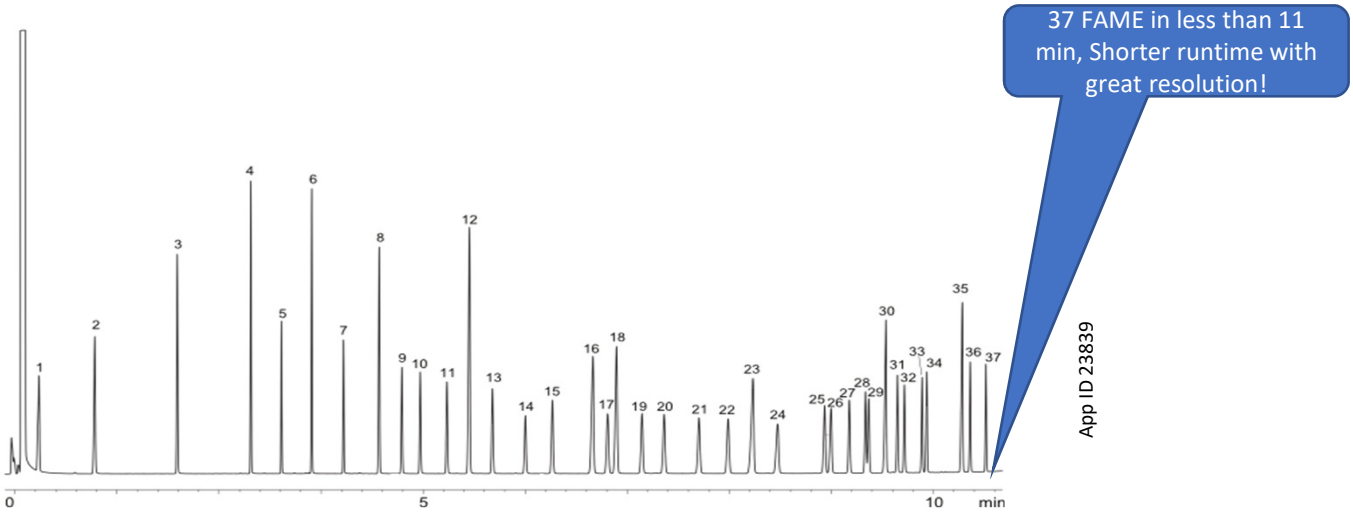
Figure 4: Separation of 37 Component FAME Mix on a 30-meter ZB-FAME GC Column



GC-FID Conditions

Column	Zebron ZB-FAME
Dimension:	30 meter x 0.25 mm x 0.20 µm
Part No.:	7HG-G033-10
Recommended Z-Guard™:	7AG-G000-00-GZK
Injection:	Split (50:1) @ 240 °C, 1 µL
Recommended Liner:	Zebron PLUS Liner Compatible with Agilent GC Instrument, Single Taper with Wool
Liner Part No.:	AG2-0A11-05
Carrier Gas:	Hydrogen @ 1.2 mL/min (constant flow)
Oven Program:	100 °C for 2 min, 140 °C @ 10 °C/min, 190 °C @ 3 °C/min, 260 °C @ 30°C/min for 2 min
Detector:	FID
Temperature:	260 °C
Analytes:	see details in Table 1

Figure 5: Separation of 37 Component FAME Mix on a 20-meter ZB-FAME GC Column



Column	Zebron ZB-FAME
Dimension:	20 meter x 0.18 mm x 0.15 µm
Part No.:	7FD-G033-05
Recommended Z-Guard™:	7AG-G000-00-GZK
Injection:	Split (100:1) @ 250 °C, 1 µL
Recommended Liner:	Zebron PLUS Liner Compatible with Agilent GC Instrument, Single Taper with Wool
Liner Part No.:	AG2-0A11-05
Carrier Gas:	Helium @ 1.0 mL/min (constant flow)
Oven Program:	80 °C for 1.5 min, 160 °C @ 40 °C/min, 185 °C @ 5 °C/min, 260 °C @ 30°C/min
Detector:	FID
Temperature:	260 °C
Analytes:	see details in Table 1

Conclusion

Zebron™ ZB-FAME GC column provides optimal separation of cis/trans FAME components. Due to extensive crosslinking through ESC™ process, this column can withstand higher temperature limit of 280 °C and provide reproducible retention of FAME compounds. ZB-FAME is available in traditional 100 meter dimension as well as in fast analysis dimensions for reproducible FAME profiling.

Need a different column size or sample preparation format?

No problem! We have a majority of our available dimensions up on www.phenomenex.com, but if you can't find what you need right away, our super helpful Technical Specialists can guide you to the solution via our online chat portal www.phenomenex.com/LiveChat.

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