Screen Beer by GC/MS Static Headspace with the Agilent J&W DB-624 Ultra Inert Capillary Column

Application Note

Food Testing & Agriculture

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Abstract

This application note highlights the utility of Agilent J&W DB-624UI columns for screening select beers by static headspace GC/MS. The inertness of the column delivers excellent peak shape for active aldehyde and organic acid analytes in complex beer matrices. Clear differences and some common elements are observable between the styles of beer investigated. The inertness and selectivity of the DB-624UI column helps to make beer profiling by static head space GC straightforward. Superior organic acid performance of the DB-624UI column is shown versus a premium competitor 624 column.

Agilent Technologies

Introduction

Small batch brewing is becoming an increasingly popular means of producing hand crafted beers and ales that find a ready market for consumers with discriminating tastes [1]. Profiling some of the flavor elements found in these beverages can help track completion of the fermentation process, assess batch quality, or evaluate the impact new or traditional ingredients have on the bouquet of flavors present in these beverages [2,3]. This work uses a highly inert DB-624UI capillary GC column to examine the constituents in several beers.

Fusel oils and related fermentation products play important roles in defining the aroma and flavor characteristics of alcoholic beverages. Fusel oils (or higher alcohols), their esters, vicinal diketones, aldehydes, and organic acids all have an effect on the balance of flavor characteristics present in a beverage. Headspace GC/MS profiling can be used to monitor the rise of desired characteristics in a batch to control off flavor elements or as a research and development tool to explore the use of new ingredients that enhance desirable taste elements in a complex matrix.

A convenient way to analyze a beer's aromatic profile is by static headspace GC/MS. Beer samples can typically be analyzed neat in a headspace vial up to an ethanol content of approximately 10% by volume. Higher ethanol content may require sample dilution to resolve peaks eluting closely with ethanol.

Experimental

An Agilent 7890/5975C GC/MS system, equipped with a split/splitless inlet, an Agilent MSD triple axis detector, Agilent 7697A Headspace Sampler and Agilent MSD Chem Station E.02.02 software, was used for GC/MS experiments. FID experiments were done using a second Agilent 7890 GC equipped with an Agilent 7693 Autosampler, and Agilent GC ChemStation B.04.03 software.

Chromatographic conditions for GC/MS headspace analysis

Column:	Agilent J&W DB-624UI, 30 m × 0.25 mm, 1.4 µm (p/n: 122-1334UI)
Carrier:	Helium, 1.8 mL/min constant flow set a 35 °C
Oven:	35 °C (5.66 min), then 8.8 °C/min to 100 °C (1.70 min), then 13.3 °C/min to 220 °C (3.39 min), then 22.1 °C/min to 250 °C (3.43 min)
Inlet:	Split/splitless, 220 °C, 1 µL split 5:1
Sample volume:	1 mL
Inlet liner:	1 mm straight single taper Ultra Inert liner (p/n: 5190-4047)
MSD:	Scan mode 30-400 amu source temperature 230 °C Quad temperature 150 °C transfer line temperature 260 °C
GC/MS:	Agilent 7890/5975C Series GC equipped with MMI and FID
Sampler:	Agilent 7697A Headspace Sampler with 111 position tray

Chromatographic conditions GC FID analysis (translated conditions)

Column:	Agilent J&W DB-624UI, 30 m × 0.32 mm, 1.8 µm (p/n: 123-1334UI)
Carrier:	Helium, 2.6 mL/min constant flow set a 35 °C
Oven:	35 °C (7.45 min), then 6.7 °C/min to 100 °C (2.23 min), then 10.1 °C/min to 220 °C (4.47 min), then 16.8 °C/min to 250 °C (4.17 min)
Inlet:	Split/splitless, 220 °C, 1 µL split 20:1
Syringe:	5 µL tapered (p/n: G4513-80206)
Inlet liner:	Ultra Inert liner with wool (p/n: 5190-3165)
GC:	Agilent 7890 Series GC equipped with a split/spiltless inlet and FID
Sampler:	Agilent 7693 Autosampler

Flow path supplies

Vials:	20 mL flat bottom crimp cap headspace vials (100 pk, p/n: 5182-0837)
Vial caps:	Headspace crimp cap /high performance septa (100 pk, p/n: 5190-3987)
Crimper:	20 mm electronic crimper (p/n: 5190-3189)
Transfer line:	0.53 mm deactivated fused silica (5 m, p/n: 160-2535-5)
Fitting:	1/6 to 1/32 in reducing fitting (p/n 0100-2594)
Septum:	Nonstick bleed and temperature optimized (50 pk, p/n: 5183-4757)
Inlet liner:	1 mm straight single taper Ultra Inert liner (p/n: 5190-4047)
Gold seal:	Gold-plated inlet seal with washer (10/pk, p/n: 5190-2209)
Ferrules:	0.5 mm id short; 85/15 Vespel/graphite (10 pk, p/n: 5062-3514)
Magnifier:	20× Magnifier loop (p/n: 430-1020)

Sample preparation

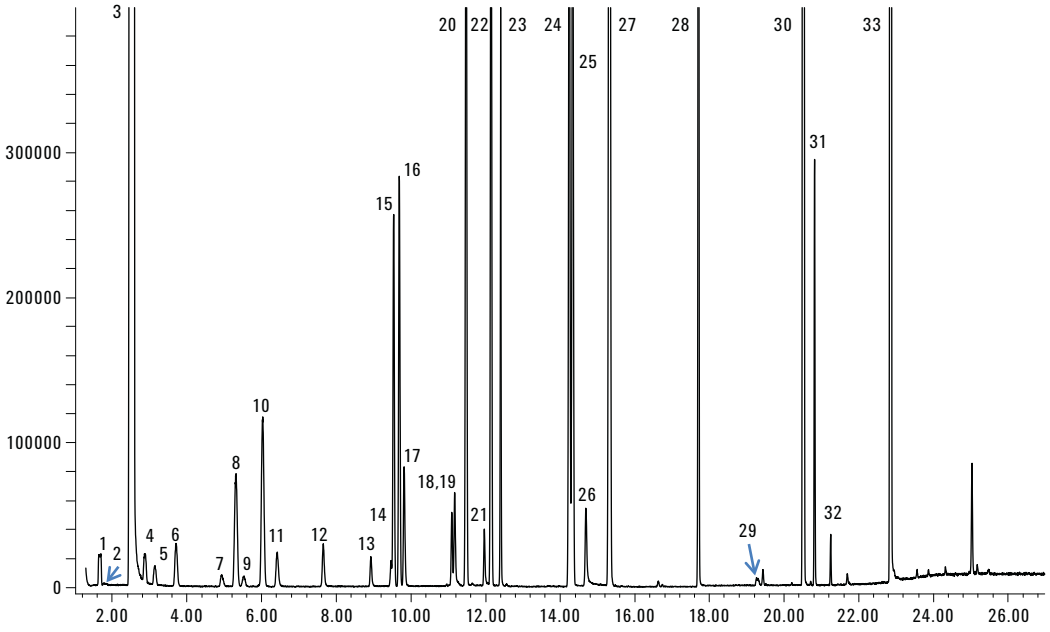
Fermentation-related alcohols, aldehydes, acetates, and organic acids were purchased from Sigma Aldrich, St Louis, MO, USA. These standards were made into stock solutions at a concentration of 1000 µL/L in ethanol (200 proof molecular biology grade purchased from Sigma Aldrich). The stock acid solution was prepared in a 5 % ethanol/water solution. Subsequent dilutions were made in deionized water.

Beer samples were purchased from a local retailer. Pale ale and a Pilsner style light beer were used for profiling by headspace GC/MS. A 10 mL aliquot of each sample was added to 20 mL headspace vials for analysis without dilution. Beer samples were allowed to equilibrate to room temperature prior to liquid transfer with a repeater pipet. A slow draw into and out of the pipet was necessary to transfer the beer without foaming.

Results and Discussion

Figure 1 shows the combined total ion chromatogram for aldehyde, fusel alcohol, and fusel acetate standard mixes at 2 µL/L. At this level, using SCAN mode, each of the standards gave high quality matches versus the National Institute of Standards and Technology (NIST) spectral library. Peaks were well resolved on a 30 m x 0.25 mm x 1.4 µm DB-624UI column. Peak shapes for aldehydes, alcohols, and esters were sharp and well defined, indicative of the highly inert character of the column.

Selectivity for the analytes of interest in the standard mix was excellent. Using a 30 m x 0.25 mm, 1.4 µm DB-624UI column, clear separation was observed between the positional isomeric pair, isoamyl alcohol, and active amyl alcohol, and also their esters. Typically, to achieve this level of separation, a 60 m column is used that requires additional run time. Here the entire run was completed in 32 minutes.



1 Acetyl aldehyde
2 Methanol
3 Ethanol
4 Acetone
5 Isopropanol
6 *tert* Butanol
7 1-Propanol
8 Butyraldehyde
9 2,3 Butanedione
10 Ethyl acetate
11 2-Butanol
12 Isobutanol
13 1-Butanol
14 2,3 Pentanedione
15 Ethyl propionate
16 Propyl acetate
17 3-Pentanol
18 Isoamyl acetate
19 Active amyl alcohol
20 Isobutyl acetate
21 1-Pentanol
22 Ethyl butanoate
23 Hexanol
24 Isoamyl acetate
25 Active amyl acetate
26 1-Hexanol
27 Heptaldehyde
28 Octanal
29 1-Phenyl ethanol
30 Ethyl octanoate
31 α-Methybenzyl acetate
32 m-Anisaldehyde
33 Ethyl decanoate

Figure 1. Total ion chromatogram of aldehyde, fusel alcohol, and fusel acetate combined standard on an Agilent J&W DB-624UI, 30 m x 0.25 mm, 1.4 µm GC column.

Screening for fermentation- and distillation-related flavor components was straightforward at the 2 µL/L level using SCAN mode. Compounds of interest eluting close to ethanol were resolvable and easily identified through NIST library matching. Lower level detection using either simultaneous SIM/SCAN or SIM modes is a very reasonable expectation for a defined set of target components with known fragmentation patterns to specify qualifying and quantifying ions.

A representative total ion chromatogram of a pale ale sample is shown in Figure 2. Several of the components contained in the standard mix are present as are some additional peaks, most notably trace amounts of ethyl hexanoate, 1-phenyl ethanol, ethyl octanoate, octanoic acid, and ethyl decanoate. There is no evidence of the peak tailing often observed when analyzing reactive compounds such as aldehydes and organic acids.

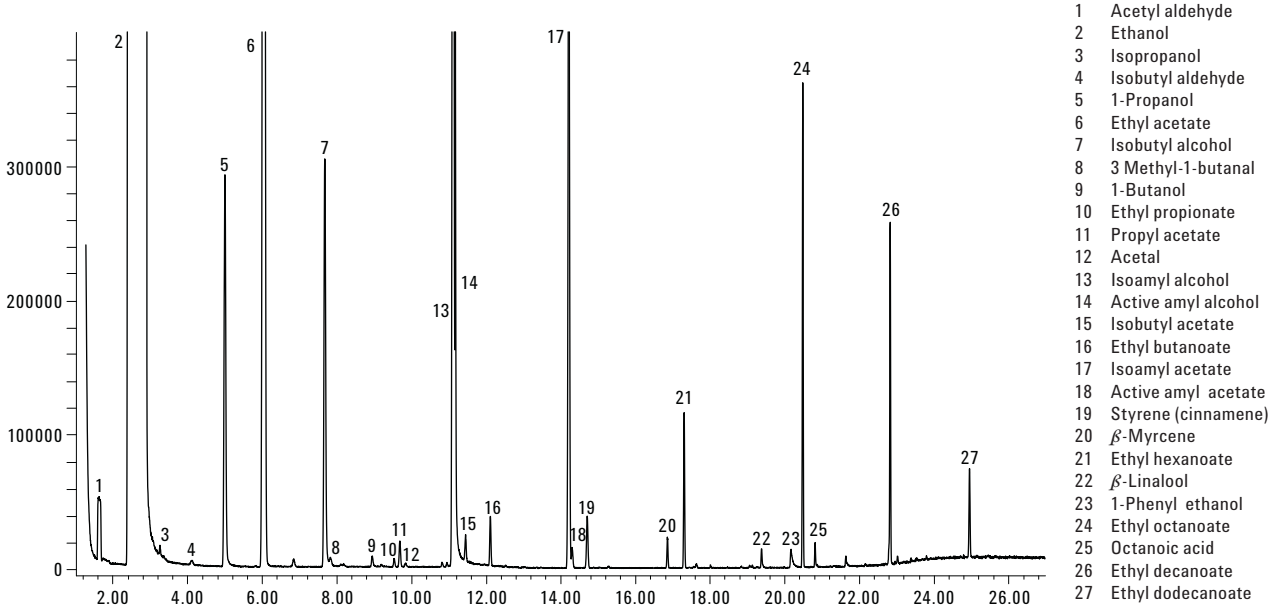


Figure 2. The total ion chromatogram of a 10 mL neat pale ale sample in the headspace vial on an Agilent J&W DB-624UI, 30 m x 0.25 mm, 1.4 µm GC column.

The total ion chromatogram of lager-style beer displays a good screening profile for the spirit in Figure 3, with observable distinctions from the pale ale profile shown in Figure 2. The lager-style beer trace is less complex than that of the pale ale sample. Acetyl aldehyde, ethyl acetate, isoamyl alcohol, active amyl alcohol, isoamyl acetate, active amyl acetate, ethyl butanoate, ethyl hexanoate, ethyl octanoate, and octanoic acid are all components found in both samples.

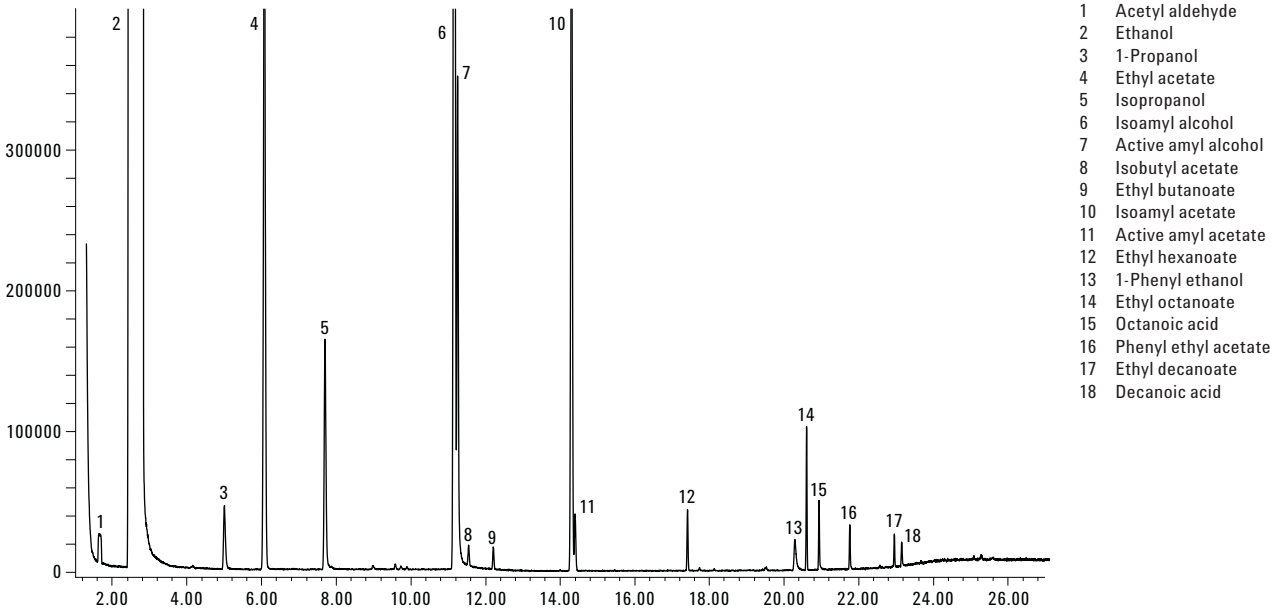


Figure 3. Total ion chromatogram of a neat lager beer sample added to in a 20 mL headspace vial on an Agilent J&W DB-624UI, 30 m x 0.25 mm, 1.4 µm GC column.

The organic acids did not transfer well into the vapor phase. For the most part, they remained in the aqueous phase. A liquid injection loading study was conducted that yielded consistent results on the range studies, 0.125 – 1 ng on-column for each component. An overlay of the FID traces for the loading injection is shown in Figure 4.

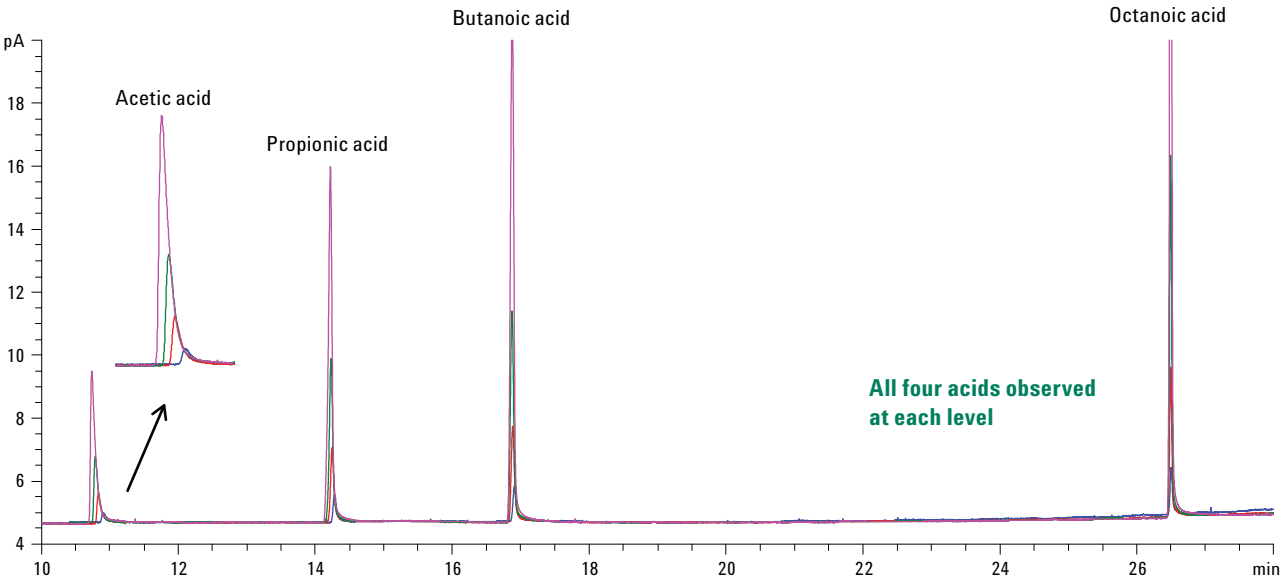


Figure 4. Overlay of the FID traces from the organic acid loading study on an Agilent J&W DB-624UI, 30 m × 0.32 mm, 1.8 µm GC column.

Organic acid recovery and peak shapes have historically been problematic for 6% cyanopropyl phenyl phases. Figure 5 shows a comparison between a DB-624UI GC column and a competitor's premium-priced 624 traditional column. Note that on the competitor's column, only 3 of the 4 organic acids investigated were detectable at the high loading level of 1 ng on-column. The DB-624UI also gave dramatically improved peak shapes for all 4 of the acids in comparison to the competitor's product.

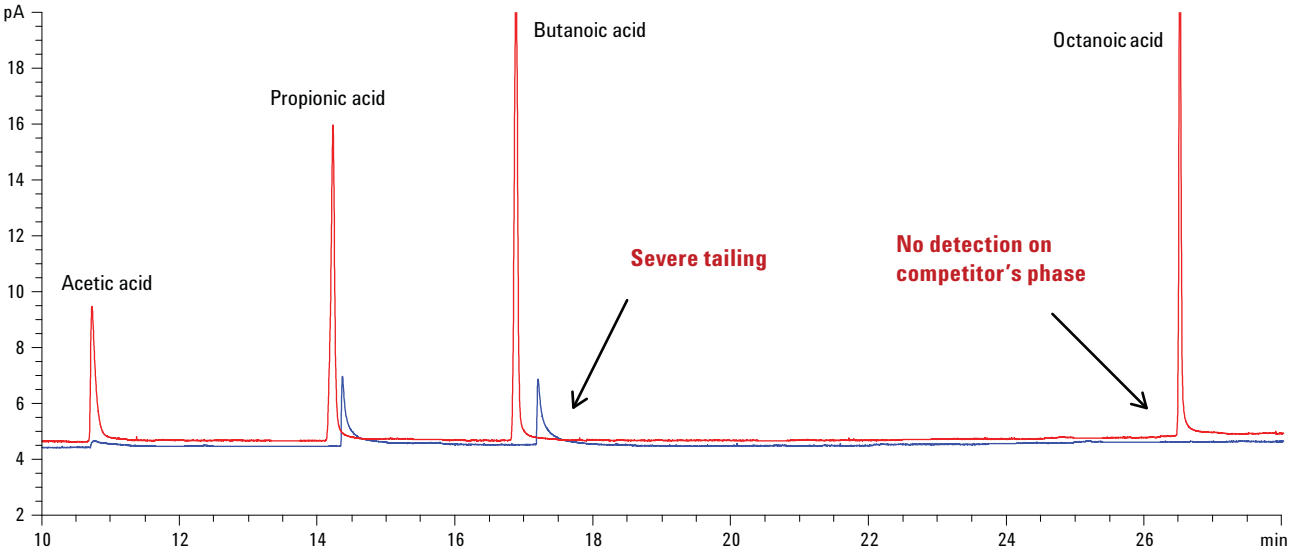


Figure 5. FID traces of a 1 ng/acid component on an Agilent J&W DB-624UI, 30 m x 0.32 mm, 1.8 µm column and a competitor's traditional 624 phase.

A 0.125 ng/acid component chromatogram is shown in Figure 6. Note that all 4 acids are detectable at this level, 8 times lower than the FID trace in Figure 5, where a competitor's premium priced traditional 624 column detects only 3 of the 4 acids with severely tailing peaks. Figure 6 shows the acid peaks are sharp and nearly symmetrical.

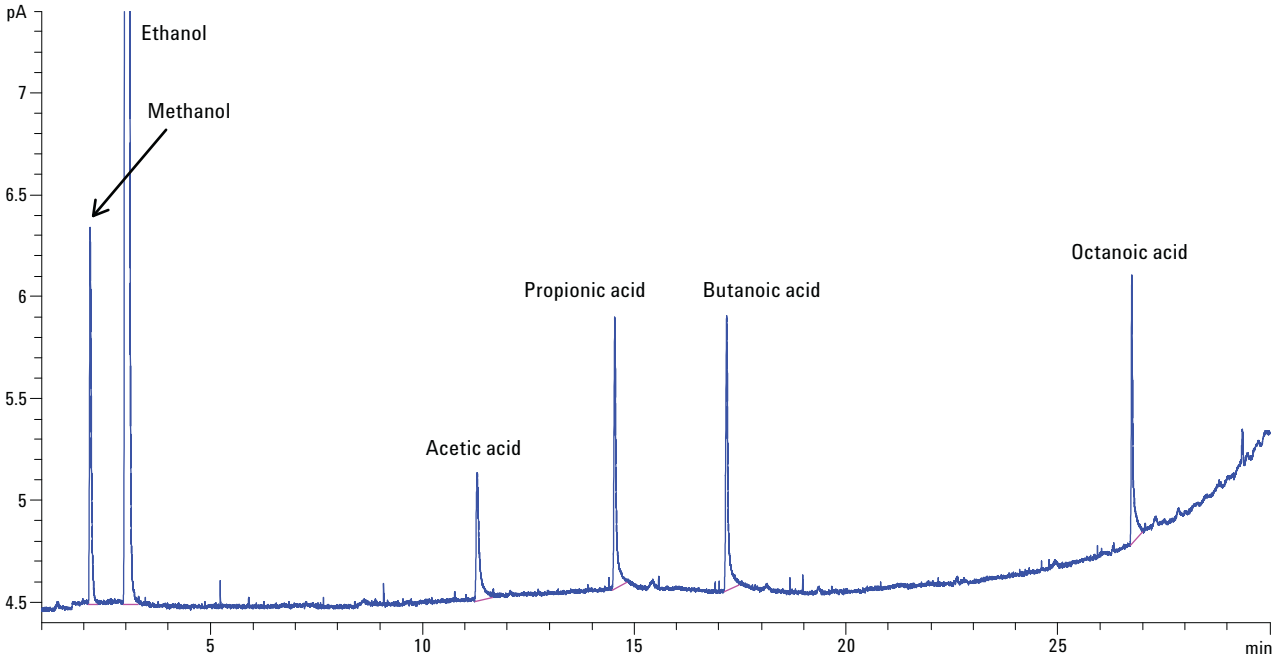


Figure 6. Highlights the organic acid performance of an Agilent J&W DB-624UI, 30 m x 0.32 mm, 1.8 µm GC column at 0.125 ng/acid component loading level. Peaks shapes are sharp even at this level.

Conclusion

The Agilent J&W DB-624UI, 30 m x 0.25 mm, 1.4 µm and 30 m x 0.32 mm, 1.8 µm GC columns deliver excellent inertness and selectivity for analytes related to fermentation and distillation in complex beer matrices. The inertness of these columns is clearly demonstrated by the sharp symmetrical peaks observed for aldehyde and alcohol components in the 2 µL/L standard and the acid components in the beer samples.

Organic acid performance for the J&W DB-624UI column is superior to a competitor's premium 624 traditional offering. The clear choice for determination of organic acids on a selective 624 phase is the J&W DB-624UI column.

References

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