Semi-Volatile Organic Compounds Analysis using an Agilent J&W DB-5 ms Ultra Inert Column

Application Note

Environmental

Authors

Yun Zou

Agilent Technologies (Shanghai) Co.Ltd

412 YingLun Road

Waigaoqiao Free Trade Zone

Shanghai 200131 P.R.China

Chongtian Yu

Agilent Technologies Co., Ltd.

11/F Cross Tower

318 Fu Zhou Road

Shanghai 200001

China

Abstract

The analysis of basic or acidic semi-volatile environmental pollutants at low concentrations places significant demands on the analytical instrument and gas chromatographic column. This application is based on EPA Method 525.2 using Agilent J&W DB-5 ms Ultra Inert columns. The ultra inert columns show exceptionally low column bleed and outstanding column inertness. By maintaining the same selectivity as DB-5 ms columns, the DB-5 ms Ultra Inert columns can be used for the existing method without method redevelopment.

Agilent Technologies

Introduction

Analysis of low levels of semivolatiles, especially active compounds, by GC or GC/MSD continues to be challenging in environmental analysis. The United States Environmental Protection Agency (US EPA) has published numerous GC/MSD methods for organic analytes in various matrices. EPA Method 525.2 is one, which is specified for drinking water [1]. The extensive analyte list includes acids, bases and neutrals. Since many of these compounds are at trace levels, the extreme sensitivity requirements make EPA method 525.2 one of the most challenging applications. Sensitivity and linearity are two areas where laboratories are seeking improved performance. Some previous publications have discussed how to lower detection limits and improve linearity by changing instrument configuration, such as using a programmable temperature vaporizing (PTV) inlet, 6-mm drawout lens, trace ion detection (TID), and a triple-axis detector (TAD) with the MSD operating in SIM mode [2, 3]. In this application, we will discuss the impact of GC column performance on sensitivity.

Improved sensitivity or maximized signal-to-noise ratio can be achieved by either reducing the background noise or increasing the signal. Over the years, GC column manufacturers have developed low bleed columns to reduce background noise level. The signal can be increased by ensuring an inert sample flow path, which will reduce peak tailing. An inert sample flow path includes liner and seal in a split/splitless inlet, the capillary column, and any metal detector parts. Peak tailing results from an interaction of active compounds with active sites within the flow path. One of the major sources of active sites is the capillary column due to its large surface area. If the number of column active sites can be reduced, higher sensitivity should be achieved. Selecting low column bleed and a highly inert column for semivolatiles analysis is very important to improve sensitivity and to ensure accurate quantitation.

The recently introduced Agilent J&W DB-5 ms Ultra Inert GC capillary column is a new generation of 5-ms columns that exhibit the highest column inertness in the industry with exceptionally low column bleed. The improved inertness performance makes DB-5 ms Ultra Inert columns especially suitable for active compounds, trace level samples, and screening of unknown sample analyses.

Agilent J&W DB-5 ms was specified in the EPA Method 525.2 and was used in this study. In addition, the Agilent J&W DB-5 ms Ultra Inert column was used since it has the same selectivity as the Agilent J&W DB-5ms. Therefore, no method

redevelopment was needed. In addition, a direct comparison of column performance between Agilent columns and that from another GC column manufacturer was performed in this study.

Experimental

Chromatographic Conditions

The experiments were performed on an Agilent 6890N Network GC System equipped with an Agilent 5975C Series GC/MSD, and an Agilent 7683 Automatic Liquid Sampler (ALS). The split/splitless inlets were fitted with long-lifetime septa (Agilent p/n 5183-4761) and splitless deactivated dual taper direct connect liners (Agilent p/n G1544-80700). Injections were made using a 5-µL syringe (Agilent p/n 5181-1273).

Table 1.   Gas Chromatography and Mass Spectrometer Conditions

GC Conditions

Column:	Agilent J&W DB-5 ms Ultra Inert, 30 m × 0.25 mm, 0.25 µm (Agilent p/n. 122-5532UI) or Agilent J&W DB-5 ms, 30 m × 0.25 mm, 0.25 µm (Agilent p/n 122-5532)
Carrier gas:	Helium, constant flow mode, 32 cm/s at 40 °C
Inlet:	Pulsed splitless at 250 °C; Purge flow 30 mL/min at 1.00 min
Oven temperature:	40 °C (1 min); 30 °C/min to 160 °C; 6 °C/min to 300 °C (5 min); Postrun: 320 °C hold 5 min
Injection size:	1 µL

MS Conditions

MS:	EI, Scan
Solvent delay:	4.0 min
MS temperature:	300 °C (Source); 180 °C (Quad)
Transfer line:	280 °C
Scan mode:	Mass range (45–450 amu)
MSD:	SIM
AutoSIM was used to pick ions, groups, and switching times	
Number of groups:	27
Compounds/group:	Varied 1 to 22
Ions/group:	Varied 2 to 55
Dwell times:	Varied 5 to 10 ms
Cycles/peak:	Minimum 10
Tune:	DFTPP

Both a multiramp temperature program and a single ramp linear temperature program were suggested in EPA method 525.2. This application used the single ramp linear temperature program, since the final oven temperature of 300 °C gave better resolution of peaks than 320 °C, which was specified in the method. Therefore, the new parameters were used for this application to test the columns, and the instrument operating parameters are listed in Table 1.

Pulsed splitless injection was used to minimize residence times of analytes in the liner, thereby reducing loss of active compounds. The inlet liner (Agilent p/n. G1544-80700) was chosen because it has shown the best performance for active compounds at low levels. It does not contain glass wool, which minimizes its contribution to the active compound degradation. This liner connects directly to the column and has a tapered top, limiting contact with metal in the inlet. Other liners can also be used, and a detailed discussion of the liners is available [4].

Full scan data acquisition was mandated by EPA Method 525. The Agilent 6890N/5975C inert GC/MSD can acquire both selected ion monitoring (SIM) and scan data in the same run. With a defined compound list, quantitation (target) ions and qualifier ions/ratios are known or can be easily determined. The use of SIM typically results in a sensitivity increase of 10 to 100 times compared to scan [5].

A source temperature of 300 °C was used instead of the typical 230 °C to 250 °C range. The higher temperature was chosen to minimize peak tailing, and therefore increase sensitivity for PAHs [6]. Lower source temperatures have historically been used to maintain performance of the active pesticides.

The system was retention time locked (RTL) to phenanthrene-d10. The primary benefit of RTL is the ability to maintain retention times after clipping or changing the column. Quantitation database and integration event times do not have to be changed. For laboratories performing SIM analyses, switching group times remain constant [7].

The column performance for EPA525.2 method was evaluated using the same instrument system and chromatographic conditions as in Table 1. Experiments were conducted on an Agilent J&W DB-5 ms column, an Agilent J&W DB-5 ms Ultra Inert column, and columns from other manufacturers respectively in both the SIM and scan modes.

Standards

EPA 525 Semivolatile Calibration Mix (1000 µg/mL, except pentachlorophenol at 4000 µg/mL), PCB Mix (500 µg/mL), EPA 525 Fortification Solution (500 µg/mL) and EPA 525 internal Standard Mix (500 µg/mL) were obtained from SUPELCO.

Pesticide samples were purchased from China National Standards Research Center at 100 µg/mL in different solvents.

Standards were made into concentrations of 0.1 to 10 ppm in dichloromethane, containing 5 ppm of ISTDs (Acenaphthylene-d10, Phenanthrene-d10 and Chrysene-d12) and SSs (1,3 Dimethyl-2-nitrobenzene, Pyrene-d10, Triphenylphosphate, Perylene-d12). Pentachlorophenol is present at four times the other analyte concentrations, as described in EPA 525.2.

Results and Discussion

Performance comparison between DB-5 ms and DB-5 ms Ultra Inert

The 10 overlaid chromatograms for the semivolatile standards are shown in Figure 1. As illustrated in Table 2, the RSD% is less than 0.098% when comparing the retention times for all 50 standards on these two columns. This same selectivity allows for direct replacement of the DB-5 ms Ultra Inert column to any existing methods developed with the DB-5 ms columns.

However, for resolution and peak height of most of semivolatile standards, slightly better performance was achieved with the DB-5 ms Ultra Inert column compared to the DB-5 ms (Figure 2). According to EPA Method 525.2, the system must provide baseline resolution of phenanthrene and anthracene; and benz[a]anthracene and chrysene should be separated by a valley whose height is less than 25% of the average peak height of these two compounds. They are closely eluting compounds with the same quantitation ions. As shown in Figure 2, the phenanthrene and anthracene pair and the benz[a]anthracene and chrysene pair were 100% baseline resolved on the DB-5 ms and DB-5 ms Ultra Inert columns, respectively. A measurement of the peak tailing was done using the US Pharmacopeia tailing factor (T_f). This is calculated using the following formula [8].

$$T_f = W5.0 / (Tw \times 2)$$

where: Tw = distance between peak front and retention time of peak (T_R) at 5% of peak height, units are the same as used for W5.0 .
W5.0 = width at 5% of height

Tailing factors (T_f) of these compounds listed in Figure 2 were close to 1. The closer the T_f value is to 1, the more symmetrical a peak. The tailing factors (T_f) could be used to evaluate the inertness performance of the columns, because column

activity leads to poor peak shapes, (tailing), poor quantification, and in extreme cases complete disappearance of active analytes at low levels. Pentachlorophenol is an aggressive compound that often shows low response and tailing peaks through adsorption onto active sites in the flow path during analysis. However, compared to the DB-5 ms and competitive 5-ms columns, the improved inertness performance provided by the DB-5 ms Ultra Inert column resulted in the most symmetrical and sharpest peaks. This increased the response for pentachlorophenol, as shown in Figure 3.
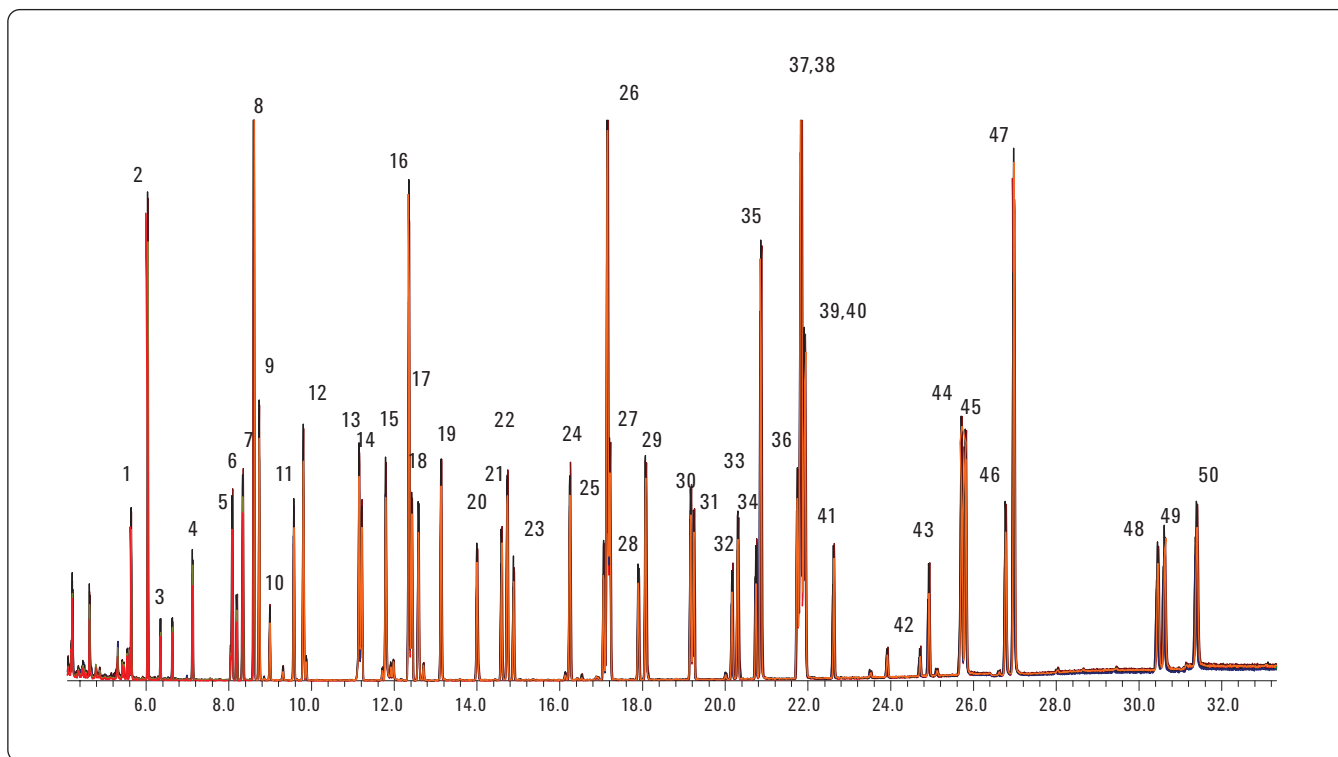

Figure 1.   10 overlaid TIC for semivolatiles, 2 ng-on column. (black-DB-5 ms Ultra Inert column, red-DB-5 ms).

Table 2. Fifty Semivolatile Compound Separation using DB-5 ms and DB-5 ms Ultra Inert Columns

Peak number	Compound	Ava. RT of DB-5 ms (n=5)	Ava. RT of DB-5 ms Ultra inert (n=5)	*RSD% of RT	Rs of DB-5 ms	Rs of DB-5 ms Ultra inert	m/z
1	Isophorone	5.647	5.642	0.068	–	–	82
2	1,3 Dimethyl-2-nitrobenzene(SS)	6.047	6.044	0.061	8.4	8.41	134
3	Dichlovos	6.351	6.349	0.059	9.05	9.03	109
4	Hexachlorocyclo-pentadiene	7.129	7.126	0.065	21.58	21.8	237
5	Dimethyl phthalate	8.084	8.087	0.071	23.14	24.1	163
6	2,6-Dinitrotoluene	8.195	8.196	0.074	2.51	2.52	165
7	Acenaphthylene	8.342	8.342	0.080	3.18	3.17	152
8	Acenaphthylene-d10(IS #1)	8.611	8.612	0.076	5.57	5.74	164
9	2-Chlorobiphenyl	8.737	8.739	0.088	2.6	2.74	188
10	2,4-Dinitrotoluene	8.993	8.996	0.085	5.44	5.6	165
11	Diethyl phthalate	9.572	9.579	0.098	12.21	12.35	149
12	Fluorene	9.804	9.808	0.088	4.65	4.6	166
13	2,3-Dichlorobiphenyl	11.153	11.159	0.095	24.87	25.63	222/152
14	Hexachlorobenzene	11.218	11.219	0.090	1.14	1.12	284
15	Pentachlorophenol	11.795	11.798	0.092	9.78	10.07	266
16	Phenanthrene-d10(IS #2)	12.357	12.363	0.088	9.28	9.34	188
17	Phenanthrene	12.426	12.432	0.091	1.16	1.13	178
18	Anthracene	12.585	12.591	0.091	2.66	2.71	178
19	2,4,5-Trichlorobiphenyl	13.133	13.140	0.089	9.27	9.61	256
20	Heptachlor	14.001	14.008	0.090	14.36	14.80	100
21	Di-n-butyl phthalate	14.587	14.600	0.095	9.99	10.14	149
22	2,2',4,4'-Tetrachlorobiphenyl	14.733	14.741	0.085	2.51	2.42	292
23	chlorpyrifos	14.882	14.892	0.088	2.50	2.53	197/97
24	2,2',3',4,6-Pentachlorobiphenyl	16.247	16.255	0.083	22.89	23.28	326
25	Butachlor	17.058	17.070	0.081	13.17	13.74	176/160
26	Pyrene-d10(SS)	17.153	17.163	0.078	1.49	1.41	212
27	Pyrene	17.214	17.223	0.076	0.87	0.91	202
28	p,p'-DDE	17.901	17.913	0.075	11.03	11.34	246
29	2,2',4,4',5,6'-Hexachlorobiphenyl	18.077	18.088	0.074	2.79	2.74	360
30	p,p'-DDD	19.162	19.176	0.075	7.54	7.04	235/165
31	o,p'-DDT	19.235	19.250	0.078	1.17	1.18	235/165
32	Benzyl butyl phthalate	20.157	20.177	0.078	14.76	15.08	149
33	p,p'-DDT	20.301	20.316	0.073	2.39	2.35	235/165
34	bis(2-Ethylhexyl)adipate	20.731	20.755	0.084	7.17	7.25	129
35	Triphenylphosphate (SS)	20.851	20.873	0.082	1.95	1.95	326/325
36	2,2'3,3',4,4'6-Heptachrolobiphenyl	21.737	21.754	0.070	13.80	13.79	394/396
37	Ben[α]anthracene	21.805	21.806	0.033	1.18	1.23	228
38	Chrysene-d12 (IS #3)	21.834	21.835	0.039	0.74	0.75	240
39	2,2',3,3',4,5,6,6'-Octachlorobiphenyl	21.903	21.904	0.036	0.94	1.04	430/428
40	Chrysene	21.928	21.928	0.034	0.80	0.80	228
41	bis(2-Ethylhexyl)phithalate	22.605	22.632	0.081	9.44	10.04	149
42	cis-Permethrine	24.692	24.720	0.077	35.23	36.04	183
43	trans-Permethrin	24.906	24.936	0.080	3.58	3.63	183
44	Benzo[b]fluoranthene	25.704	25.725	0.064	12.10	12.29	252
45	Benzo[k]fluoranthene	25.802	25.824	0.067	1.30	1.40	252
46	Benzo[a]pyrene	26.766	26.789	0.064	12.69	13.60	252
47	Perylene-d12(SS)	26.966	26.991	0.067	2.60	2.74	264
48	Indeno[1,2,3-c,d]pyrene	30.434	30.470	0.077	40.75	40.99	276
49	Dibenz[a,h]anthracene	30.594	30.635	0.086	1.69	1.76	278
50	Benzo[g,h,i]perylene	31.373	31.414	0.088	7.74	8.07	276

*RSD% was calculated by using the retention times for each compound on the DB-5 ms and DB-5 ms Ultra Inert columns.
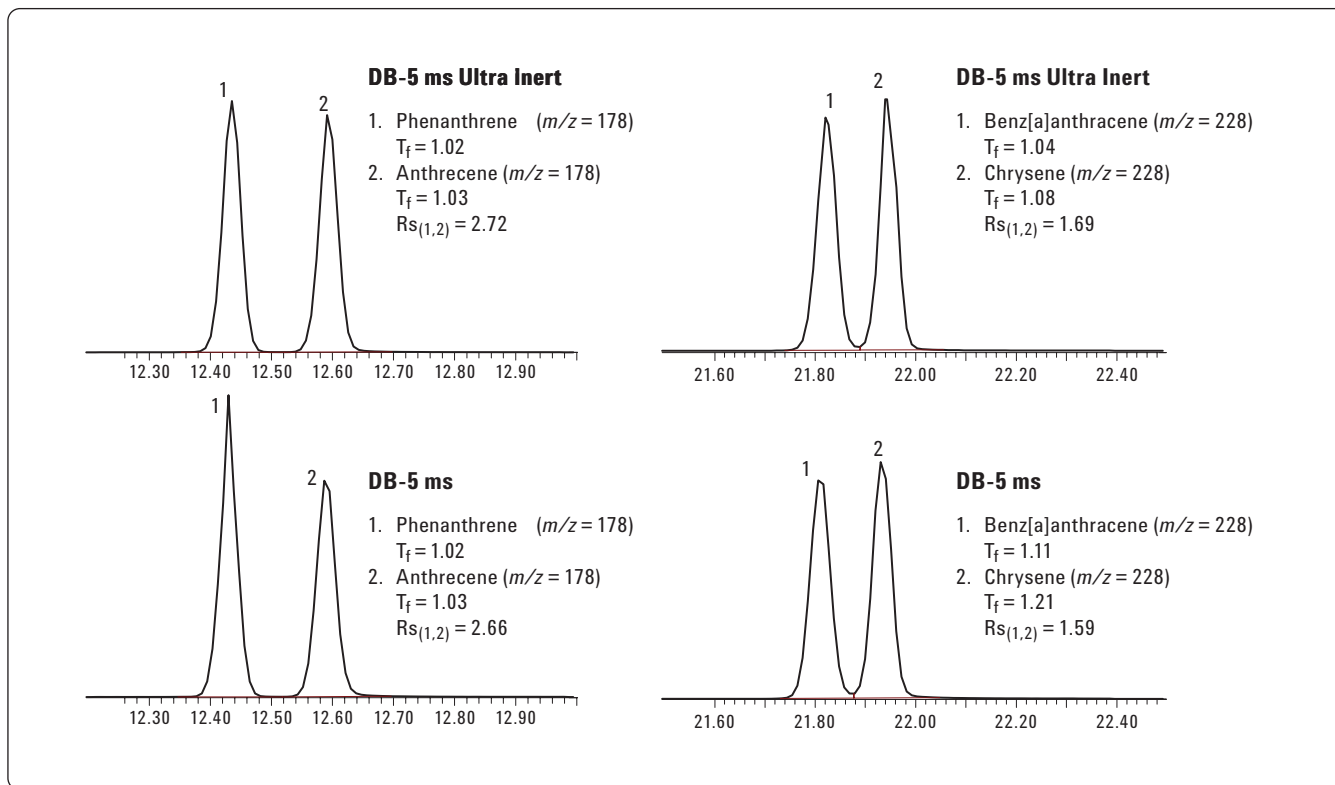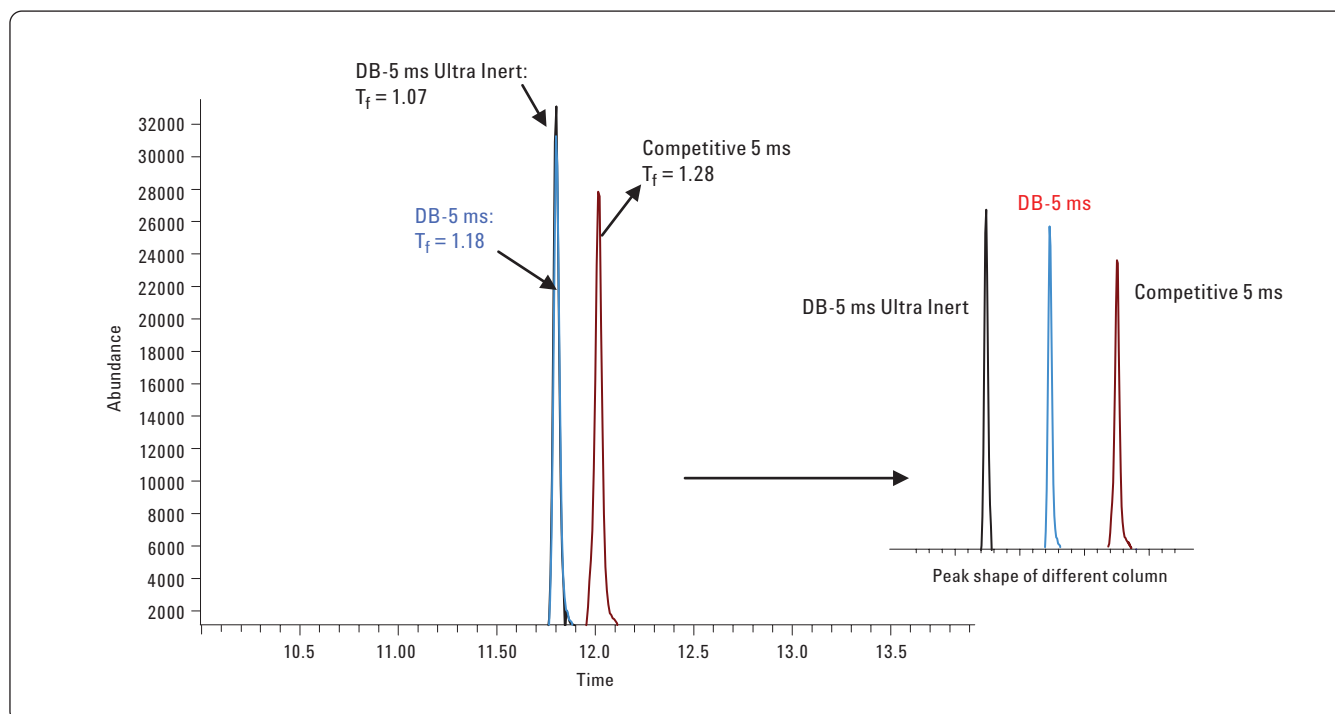
Figure 2.   Resolution of critical pairs.

DB-5 ms Ultra Inert

1. Phenanthrene   (m/z = 178)
    $T_f = 1.02$
2. Anthrecene (m/z = 178)
    $T_f = 1.03$
    $Rs_{(1,2)} = 2.72$

DB-5 ms Ultra Inert

1. Benz[a]anthracene (m/z = 228)
    $T_f = 1.04$
2. Chrysene (m/z = 228)
    $T_f = 1.08$
    $Rs_{(1,2)} = 1.69$

DB-5 ms

1. Phenanthrene   (m/z = 178)
    $T_f = 1.02$
2. Anthrecene (m/z = 178)
    $T_f = 1.03$
    $Rs_{(1,2)} = 2.66$

DB-5 ms

1. Benz[a]anthracene (m/z = 228)
    $T_f = 1.11$
2. Chrysene (m/z = 228)
    $T_f = 1.21$
    $Rs_{(1,2)} = 1.59$

DB-5 ms Ultra Inert:
$T_f = 1.07$

Competitive 5 ms
$T_f = 1.28$

DB-5 ms:
$T_f = 1.18$

DB-5 ms

DB-5 ms Ultra Inert

Competitive 5 ms

Peak shape of different column

Figure 3.   Extracted ion chromatograms of pentachlorophenol, m/z 266.

Performance Comparison Between DB-5 ms Ultra Inert and Competitive Column

A column performance comparison of the DB-5 ms Ultra Inert column and a 5-ms column from another manufacturer is shown in Figure 4. Elution order of some compounds were not the same for these two columns, such as peak 25 (butachlor), peak 36 (2,2'3,3',4,4'6-heptachrolobiphenyl) and peak 39 (2,2',3,3',4,5,6,6'-octachlorobiphenyl). As demonstrated in Figure 5, much lower column bleed was achieved on the DB-5 ms Ultra Inert column at high temperature compared to the competitive column. The lower bleed profile on the DB-5 ms Ultra Inert column provides greater signal-to-noise, lower detector maintenance, and potential for longer column life. In addition, indeno[1,2,3-c,d]pyrene (peak 48) and dibenz[a,h]anthracene (peak 49) are very difficult to baseline resolve, which was achievable on the DB-5 ms Ultra Inert column.

There are many active compounds such as pentachlorophenol and pesticides in these semi-volatile standards. As discussed

before, the peak symmetry could be used to evaluate the inertness performance of the columns. The T_f values for pentachlorophenol, dichlovos, heptachlor, chlorpyrifos and p,p'-DDE peaks are included in the chromatograms in Figure 3 and Figure 6. The DB-5 ms Ultra Inert column shows a significant reduction in the tailing factors. For example, the tailing factor of pentachlorophenol on the DB-5ms Ultra Inert column is 1.07 whereas the T_f is 1.28 for the competitive 5-ms column (Figure 3).

As shown in Figure 6, the competitive 5-ms column shows severe peak tailing, especially for dichlovos and chlorpyrifos, where the T_f values are 0.80 and 0.77, respectively. In addition, due to peak tailing, there is a noticeable loss of response with these two peaks on the competitive 5-ms column. In contrast, quite symmetrical peak shapes were obtained for dichlovos and chlorpyrifos with the DB-5 ms Ultra Inert column, with tailing factors being very close to 1. Furthermore, strong responses allowed for accurate quantitation especially at trace levels. The DB-5 ms Ultra Inert column demonstrates the benefits that more inert surfaces can have for the analysis
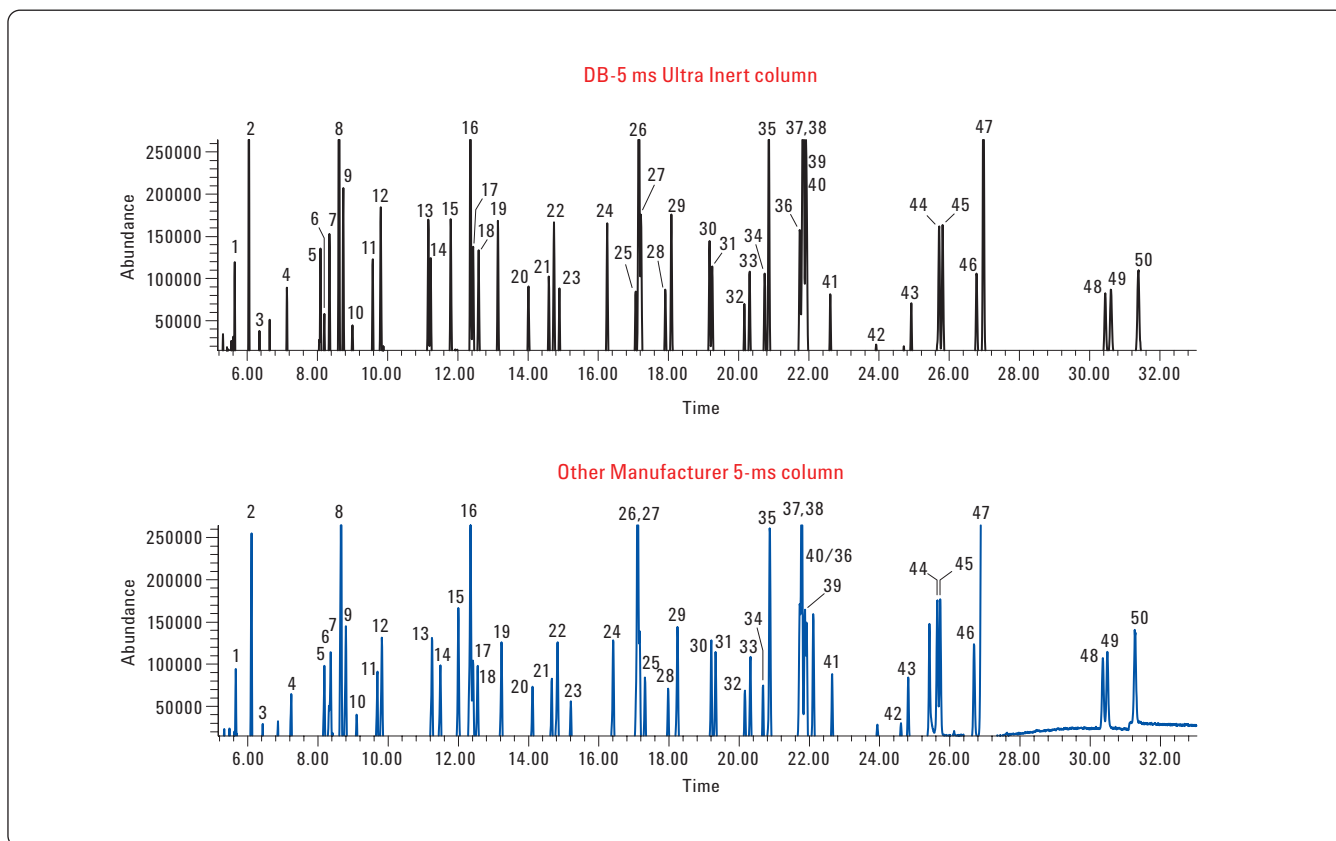

Figure 4.   Comparison of Semivolatile Standards using DB-5 ms Ultra Inert column and 5-ms column from other manufacturer, 1 ng on-column.
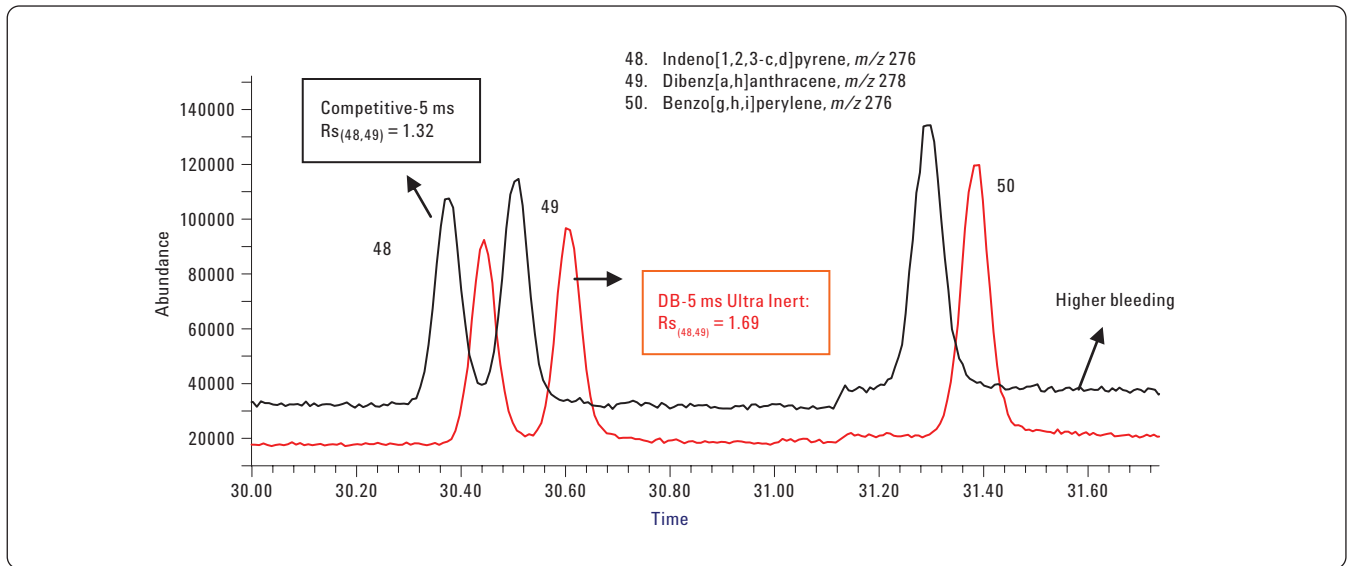
7

Figure 5.   Expanded view from Figure 3 shows lower bleed and better separation of late-eluting compounds on DB-5 ms Ultra inert column.
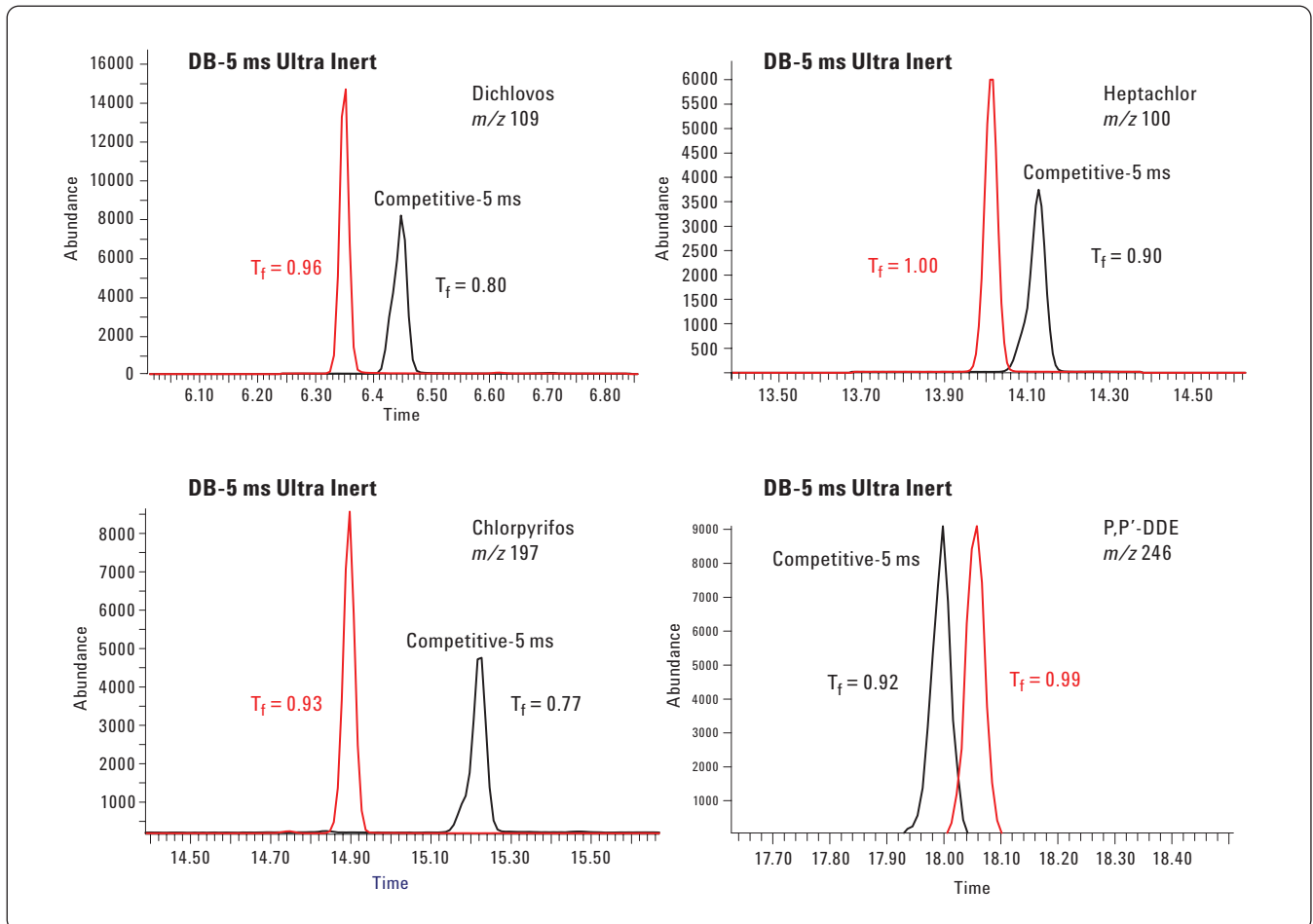

Figure 6.   Extracted ion chromatograms of pesticides.

of active compounds.

Conclusions

According to the USEPA Method 525.2 criteria, the Agilent J&W DB-5 ms Ultra Inert column can provide excellent chromatography for semivolatile compounds, including difficult to analyze active compounds at trace levels. The higher inertness and lower column bleed compared to a competitive column showed its superior performance for active compounds. In addition, the DB-5 ms Ultra Inert column has the same selectivity as the DB-5 ms column, making it very straightforward to translate any existing method from the DB-5 ms column to the DB-5 ms Ultra Inert column without method redevelopment.

References

1. USEPA Method 525.2: Methods for the Determination of Organic Compounds in Drinking Water Supplement III, National Exposure Research and Development U.S. EPA, Cincinnati, OH (August 1995).

2. M. Szelewski, "Femtogram GC/MSD Detection Limits for Environmental Semivolatiles Using a Triple-Axis Detector", Agilent Technologies, publication 5989-8342EN, www.agilent.com/chem

3. M. Szelewski, "Drinking Water Semivolatiles Analysis using the 6890N/5975B inert GC/MSD," Agilent Technologies, publication 5989-5421EN, www.agilent.com/chem.

4. M. Szelewski, B. Wilson, and P. Perkins, "Improvements in the Agilent 6890/5973 GC/MSD System for Use with USEPA Method 8270," Agilent Technologies publication 5988-3072EN, www.agilent.com/chem.

5. C. K. Meng, "Identification and Quantitation of Pesticides in the Parts-per-Trillion Range Using Retention Time Locking and GC/MS," Agilent Technologies, publication 5988-4392EN, www.agilent.com/chem.

6. M. Szelewski, "Synchronous SIM/Scan Low- Level PAH Analysis Using the Agilent Technologies 6890/5975 Inert GC/MSD," Agilent Technologies, publication 5989-4184EN, www.agilent.com/chem.

7. K. Weiner, N. Mata, and P. Wylie, "Retention Time Locking with the G1701BA MSD Productivity ChemStation," Agilent Technologies, publication 5968-3433E, www.agilent.com/chem.

8. "Understanding Your ChemStation," Agilent Technologies manual part number G2070-91125, page 246, Edition 07/08.

For More Information

For more information on our products and services, visit our Web site at www.agilent.com/chem.

www.agilent.com/chem

Agilent Technologies