Femtogram GC/MSD Detection Limits for Environmental Semivolatiles Using a Triple-Axis Detector

Application

Environmental

Author

Mike Szelewski
Agilent Technologies, Inc.
2850 Centerville Road
Wilmington, DE 19808
USA

Abstract

The analysis of semivolatiles at very low levels presents challenges due to analyte activity, background contamination, and instrument sensitivity. Method requirements vary worldwide, with the least sensitive specifying 1-µL injections and full-scan data acquisition. The lowest detection limits can be achieved using a programmable temperature vaporizing (PTV) inlet, trace ion detection (TID), and a triple-axis detector (TAD) with the MSD operating in SIM mode.

Introduction

Low-level semivolatiles analysis is used to concurrently measure a mixture of acids, bases, neutrals, and pesticides in drinking water or source water. Most laboratories analyze for > 100 compounds, with a chromatographic run time of 25 to 40 minutes. Sample extraction is accomplished using liquid-solid extraction (LSE) with C_{18} disks or cartridges. Liquid-liquid extraction with a solvent such as dichloromethane is an alternative technique. Extract injection is typically 1 µL hot splitless with the MSD operating in full-scan mode, as specified in some commonly used methods such as USEPA Method 525.2 [1].

Sensitivity is an area where laboratories are seeking improved performance; it can be affected by sample preparation, extract volume injected, instrument tuning, signal acquisition, and overall system activity. Sensitivity is also a confusing term, with all of the following used interchangeably: maximum sensitivity, minimum sensitivity, best sensitivity, lowest detection limit, instrument detection limit (IDL), and method detection limit.

Previous publications have focused on activity/linearity, speed, productivity, and large-volume injection [2–5]. Sensitivity is a factor in all of these, and many times is a trade-off.

This application addresses the parameters that affect the IDL, that is, the "sensitivity" of the GC/MSD system. There are statistical ways to calculate the IDL, but these may not answer the questions, "How much can I actually see?" or "What is the lowest amount that will produce a peak I can integrate?"

Instrument Operating Parameters

The recommended instrument operating parameters are listed in Table 1. These are starting conditions and may have to be optimized. For the best sensitivity, parameters should be chosen that transfer the maximum amount of analyte onto the column. Furthermore, the entire system must be inert, as sensitivity is almost always lost on active analytes first.

Many analysts associate the use of PTV only with large-volume injection (LVI) in solvent vent mode [4]. LVI will allow lower levels of calibration, but

Table1. Gas Chromatograph and Mass Spectrometer Conditions

GC	Agilent Technologies 7890A or 6890N		
Inlet	EPC PTV		
Mode	Splitless		
Temperature ramp	°C/min	Next °C	Hold min
Initial		20	0.05
Ramp 1	600	350	0.90
Ramp 2	10	250	0.00
Cryo	On		
Cryo use temperature	100 °C		
Cryo timeout	10.00 min (On)		
Cryo fault	On		
Pressure	11.40 psi (On)		
Purge flow	30.0 mL/min		
Purge time	1.50 min		
Total flow	34.4 mL/min		
Gas saver	Off		
Gas type	Helium		
PTV Liner	Agilent multi-baffle liner, no packing, p/n 5183-2037		
Oven	120V		
Oven ramp	°C/min	Next °C	Hold min
Initial		40	2.50
Ramp 1	50	110	0.00
Ramp 2	10	320	1.10
Total run time	26 min		
Equilibration time	0.5 min		
Oven max temperature	325 °C		
Column	Agilent Technologies HP 5 MSi, p/n 19091S-433i		
Length	30.0 m		
Diameter	0.25 mm		
Film thickness	0.25 µm		
Mode	Constant flow		
Pressure	11.40 psi		
Nominal initial flow	1.4 mL/min		
Inlet	Front		
Outlet	MSD		
Outlet pressure	Vacuum		
RTL	System retention time locked to phenanthrene-d10 at 12.700 min		

Front Injector	
Sample washes	1
Sample pumps	2
Injection volume	2.0 µL
Syringe size	10 µL
PreInj Solv A washes	0
PreInj Solv B washes	1
PostInj Solv A washes	3
PostInj Solv B washes	2
Viscosity delay	0 seconds
Plunger speed	Fast
PreInjection dwell	0 minutes
PostInjection dwell	0 minutes
MSD	Agilent Technologies 5975C, Triple-Axis Detector
Drawout lens	3 mm standard aperture drawout lens
Solvent delay	4 min
Low mass	45 amu
High mass	450 amu
Threshold	0
Sampling	2
Quad temp	180 °C
Source temp	300 °C
Transfer line temp	280 °C
Tune type	Autotune
EMV mode	Gain factor = 1

MSD-SIM

AutoSIM was used to pick ions, groups and switching times

Number of groups	25
Compounds/group	Varied 1 to 22
Ions/group	Varied 2 to 45
Dwell time, msec	Varied 5 to 50
Cycles/peak	Minimum 10

Calibration Standards

Ultra Scientific, North Kingstown, RI. p/n DWK-5252. Four mixtures, co-diluted in dichloromethane, resulting in 108 compounds at 7 concentration levels: 10, 4, 1, 0.4, 0.1, 0.04, and 0.01 ppm. Each level spiked with 3 Internal Standards at 2 ppm and 4 surrogate standards at 2 ppm. Each level then diluted 1:100 in dichloromethane, resulting in 7 concentration levels: 100, 40, 10, 4, 1, 0.4, and 0.1 ppb (pg/uL) with IS/SS at 2 ppb.

method development is necessary to optimize recovery of compounds while eliminating the solvent. LVI also injects more matrix and may not improve Signal-to-Noise (S/N) due to chemical noise. The PTV has other operating modes; "cold" splitless mode was used here. Splitless injection into a cold inlet instead of a typical hot splitless inlet offers these advantages:

1. Solvent expansion is minimized; analytes do not travel outside the liner and contact metal surfaces, thereby minimizing degradation.
2. Analytes vaporize at the lowest temperature, also minimizing degradation.
3. Volatile solvent is transferred onto the column first; analyte peak shape is improved for injections of 2 to 5 µL.

Figure 1 shows the PTV temperature and flow programs together with the oven program. The PTV is held at 20 °C, a temperature below the boiling point of the solvent dichloromethane, 39.8 °C, during the fast injection period, 0.05 min. At the end of the injection period, the PTV is rapidly heated to 350 °C, transferring analyes onto the column. At the end of the splitless time, 1.5 min, the inlet is purged at 30 mL/min. The PTV is allowed to cool during the run.

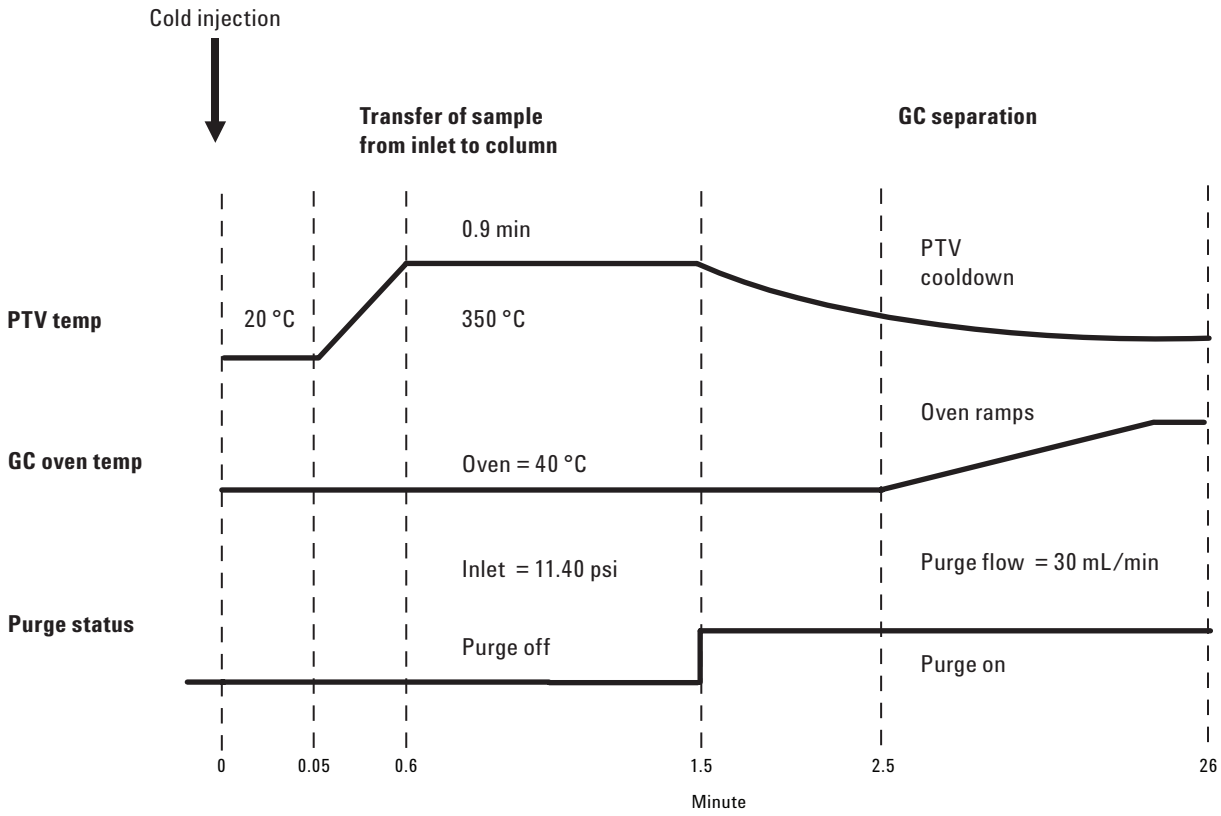

Figure 1. PTV cold splitless temperature and flow programs.

The PTV program ramp can be adjusted and multiple ramps are possible. The PTV inlet liner (p/n 5183-2037) is multi-baffled and deactivated. It does not contain glass wool, which could contribute to active compound degradation. This liner has sufficient capacity to accommodate a 2- to 5-µL injection volume at fast speed. A 2-µL injection was used for all data presented here.

The oven program relationship to the PTV parameters is shown in Figure 1. The oven starts at 40 °C and is held there during the injection cycle and splitless transfer of analytes onto the column. The oven then programs rapidly to 110 °C, followed by a slower ramp for compound separation. There is an extra 1 min of oven hold time at 40 °C, which is between 1.5 and 2.5 min. This maintains the retention time locked (RTL) times for analytes while providing room for the injection to be scaled up to LVI, if desired. The 240V oven was used, but a 120V oven can also achieve the ramp rates found in Table 1.

The HP-5MSi column is designed for inertness and is well suited to this method. This is the latest version of the most popular column in environmental laboratories, the HP-5MS. The column was run in constant-flow mode at 1.4 mL/min to maintain peak shape and sensitivity.

The system was RTLocked to phenanthrene-d10 at 12.700 min. The primary benefit of RTL for this analysis is maintaining constant switching times for SIM groups. After clipping the column, a rerun and analysis of the locking standard is all that is needed to restore shifted peak times. Quantitation database and integration events times also do not have to be changed. Additional RTL applications detailing the numerous benefits of RTL are available at www.agilent.com/chem. It is almost impossible to use a method with this many SIM groups without RTL, in a productive laboratory.

The standard 3-mm drawout lens was used for best sensitivity. Previous work has shown improved linearity across a wide calibration range using the optional 6-mm lens [1]. Using the 6-mm lens will show a typical loss of 2 to 5x in the IDL.

The 5975C MSD was equipped with a Triple-Axis Detector (TAD) [6]. The TAD presents several advantages to the user, one of which is, “Although signal is enhanced, neutral noise is substantially reduced through the off-axis design.” This increase in S/N for clean samples with minimal chemical noise can help reach a lower IDL. Trace ion detection (TID) was switched on during all data acquisition [7]. TID is a filtering routine to minimize noise and is selectable in the software.

Scan parameters are listed and data were collected in either scan mode or in SIM mode. None of the runs was made in synchronous SIM/scan mode. A sampling rate of 2 was used, as it is typical of most methods on a 250-µm id column. This sampling rate, with a 45 to 450 mass range, resulted in at least 10 scans across each peak.

AutoSIM setup was used in combination with the scan quantitation database to pick ions, groups, and switching times. The SIM acquisition table from AutoSIM was used directly with only two modifications. Tebuthiuron (ion 156) and tricyclazole (ion 189) are known for poor peak shape. Their ions were manually added to the groups across which the peaks eluted. A target ion plus one qualifier ion were used for all internal (ISTDs) and surrogate standards (SSs). A target ion plus two qualifier ions were used for all other analytes, if they were present in sufficient abundance in the spectra. A minimum of 10 SIM data points were acquired across each peak.

A source temperature of 300 °C was used instead of the typical 230 to 250 °C range. This higher temperature has been used to minimize peak tailing, and therefore improve sensitivity for PAHs [5].

The compound list was taken from USEPA 525 and is typical of the analytes that laboratories worldwide are interested in analyzing at low levels. The USEPA 8270 list was not used, as it is targeted at higher concentrations of compounds in waste samples that contain high levels of matrix and are not comparable here. The best way to improve sensitivity for solids and waste samples is through extract cleanup. The standards were prepared in dichloromethane only for the single component analytes, except disulfoton sulfoxide and disulfoton sulfone, which were not included in the commercially available mixture. Standards were not prepared for multicomponent toxaphene or the Aroclors.

A typical calibration range for low-level semivolatiles is 0.1 to 10 ppm as defined in USEPA 525. Standards were made from 0.01 to 10 ppm, containing 2 ppm of ISTDs and SSs. A dilution of 1:100 of each of these yields a range of 0.1 to 100 ppb, with ISTDs and SSs at 20 ppb, for a lower working range. Atrazine and alachlor are present in two of the stock mixes, so their concentrations are twice that of other analytes. Pentachlorophenol is present at four times the other analyte concentrations, as described in USEPA 525.

Results

The standard solutions from 0.1 to 100 ppb were run in both SIM and scan modes. Data from the 0.1-ppb scan injections showed insufficient response or were too noisy to reproducibly integrate. The SIM data at 0.1 ppb were significantly improved compared to the scan data and could be routinely used. A listing of selected analytes with S/N measured from 1.0 ppb scan runs (2 pg) are shown in Table 2, together with data from 0.1- (0.2-pg) and 1.0-ppb SIM runs. Each value is an average of three acquisitions on one system, using peak-to-peak noise.

Table 2. Signal-to-Noise for Selected Analytes, SIM and Scan Modes

Compound	Ion	pg → RT	0.2 SIM S/N	2.0 SIM S/N	2.0 Scan S/N
Hexachlorocyclopentadiene	237	7.960	6.3	77	7.5
Trifluralin	264	11.608	4.4	49	7.7
Simazine	201	12.274	1.0	16	2.4
Atrazine	200	12.385	3.1	30	13
Pentachlorophenol	266	12.492	2.4	20	3.7
Chlorothalonil	266	13.146	2.6	26	2.9
Aldrin	66	14.661	1.6	15	1.9
Heptachlor epoxide	353	15.429	6.2	49	3.4
4,4′-DDE	246	16.557	7.0	72	17
Carboxin	143	16.696	2.4	22	4.0
Endrin	263	17.003	2.3	22	4.1
4,4′-DDD	235	17.323	7.5	76	7.5
4,4′-DDT	235	18.000	5.9	60	5.9

There is excellent agreement between the SIM S/N values at the two levels for most compounds. This shows that the responses are real and that the entire system is inert. There is a slight loss of simazine and minimal interference for pentachlorophenol and heptachlor epoxide at the lowest level, 0.2 pg. At the 200 femtogram level, this is no surprise.

The scan S/N at 2.0 pg is lower than SIM, as expected, by 3- to 15-fold. The gains in S/N moving from scan to SIM are related to the dwell time versus the original sampling rate.

Extracted Ion Currents (EICs) from the 1.0-ppb level for both SIM and scan are shown in Figures 2a to 2d. It can clearly be seen that either the SIM or scan signals could be used for quantitation based on S/N and peak shape. Of particular note is the response and very good peak shape for pentachlorophenol, even at an 8-pg full scan.

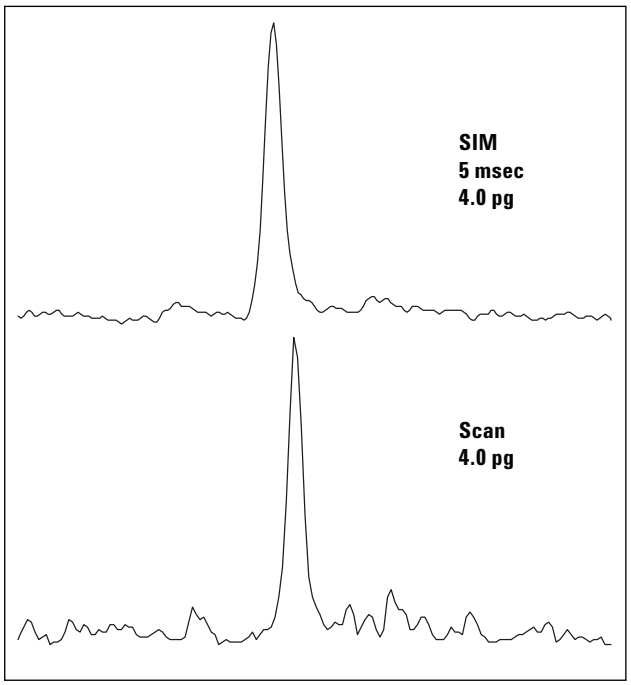

Figure 2a. Atrazine – Extracted Ion 200, RT 12.350 min.

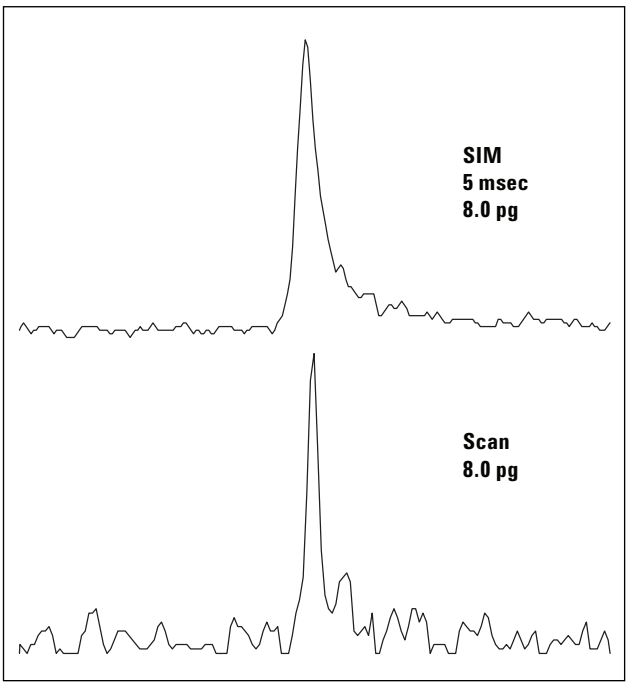

Figure 2b. Pentachlorophenol – Extracted Ion 266, RT 12.445 min.

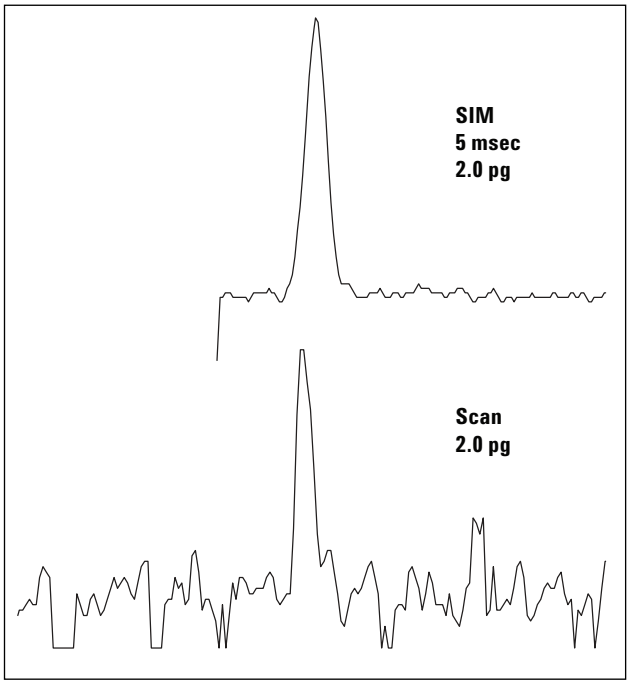

Figure 2c. Aldrin – Extracted Ion 66, RT 14.616 min.

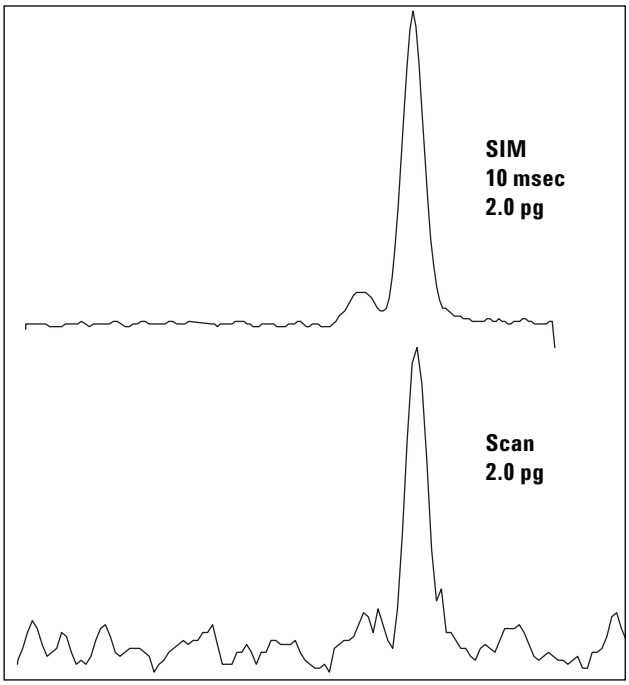

Figure 2d. 4,4′-DDT – Extracted Ion 235, RT 18.00 min.

Although linearity is not the focus of this application, it is a measure of inertness, reproducibility, and sensitivity. Linearity can be determined by the percent relative standard deviation (%RSD) of the relative response factor (RRF) for each compound across the calibration range. The %RSD and the RRF calculations are done automatically by the GC/MSD ChemStation software in conjunction with Excel. There is no correct %RSD, as it is method dependent. The %RSDs of the RRFs for selected compounds are shown in Table 3.

Table 3. Linearity of Selected Analytes

Calibration range pg →	0.2–200 SIM	2–200 Scan
Compound	**%RSD**	**%RSD**
Dichlorvos	1.9	7.0
Mevinphos	10.1	7.0
2,3-Dichlorobiphenyl	5.3	3.0
Atrazine	14.2	14.5
Pentachlorophenol	6.3	33.0
Anthracene	2.2	3.0
Chlorothalonil	7.6	25.0
Heptachlor epoxide	6.6	13.0
4,4′-DDE	4.5	9.0
4,4′-DDD	7.4	8.0
4,4′-DDT	4.0	5.9

At first glance some of the %RSD values appear high, such as pentachlorophenol (PCP) and chlorothalonil. These are calibrated, however, from 2 to 200 pg in scan mode, which is 50-fold lower than USEPA 525 mandates. The SIM data are calibrated from 0.2 to 200 pg, which is 500-fold lower and a 10-fold wider range. This demonstrates both inertness and detectability at the femtogram level.

As an additional overall measure of system linearity, the average of all %RSDs was calculated at 8% for SIM data and 13% for scan data. Not all compounds were calibrated to the 0.1-ppb level, as they did not have a signal that could be reliably measured. The phthalates, easily detected at low levels, were excluded from these averages due to common laboratory contamination.

EICs at the 200-femtogram level, from SIM, are shown for six different compounds in Figures 3. All are easily seen and measured against noise. As an analyst's measure of sensitivity, the question from the introduction was "How much can I actually see?" The answer: very low picogram levels for most environmental semivolatiles in scan mode. The IDL using SIM is even lower, in the femtogram range.

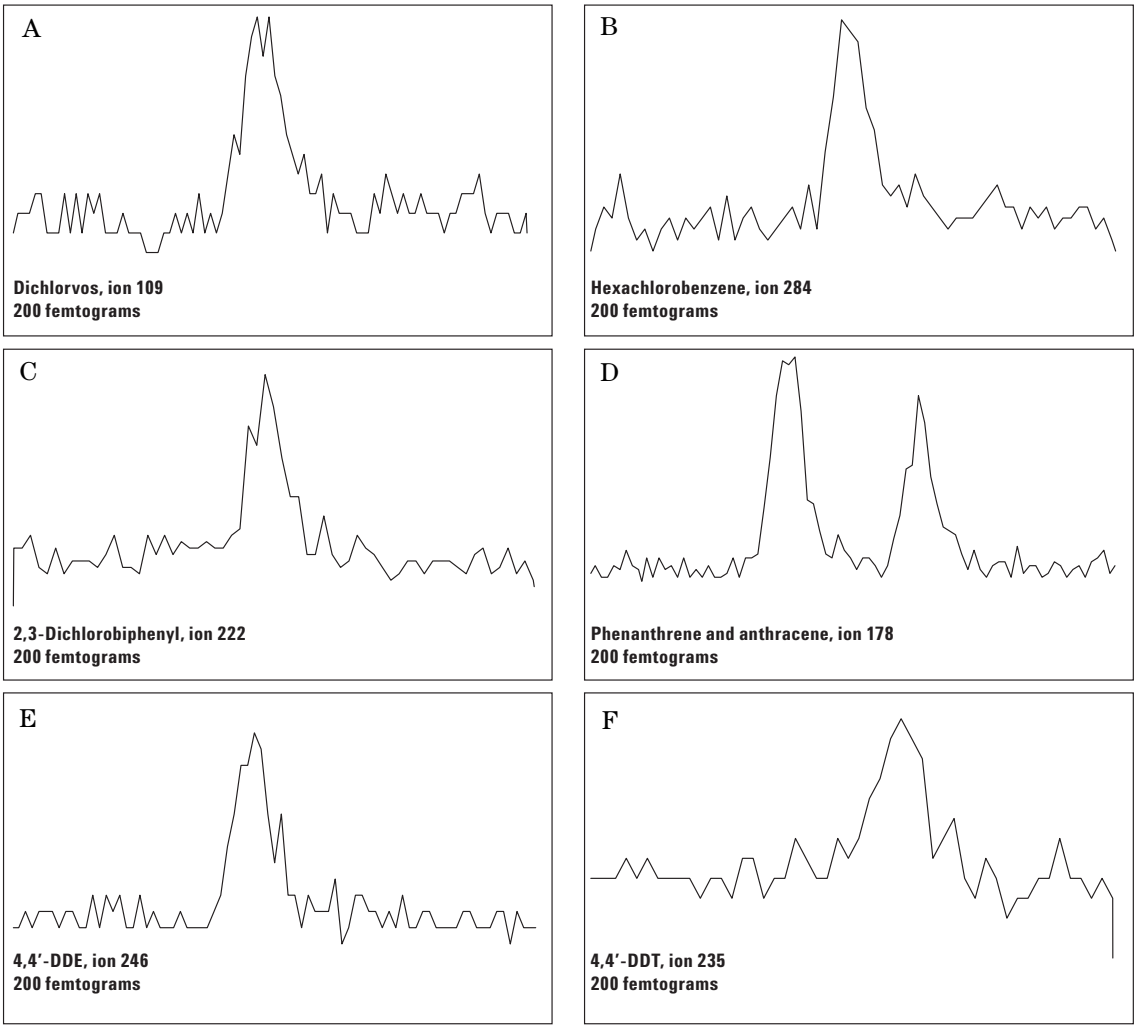

Figure 3. EICs at the 200 femtogram level.

Conclusions

Traditional semivolatiles methods can be altered to achieve better instrument detection limits. There have been advancements in hardware, such as the Triple-Axis Detector (TAD), that improve sensitivity. Signal handling using Trace Ion Detection (TID) provides better S/N through lower noise. The PTV, used in "cold" splitless mode, maximizes the amount of sample on the column, while vaporizing analytes at the lowest possible temperature. Coupled with an inert column and source, the PTV provides an easy way to improve sensitivity. Methods that require only a target ion and a few qualifier ions for identification can often be changed to SIM from scan, improving S/N by 3- to 50-fold. Combining all of these hardware, software, and operating parameters can result in femtogram instrument detection limits (IDLs) and sensitivity you can use.

References

1. USEPA Method 525.2 is available from different sources listed on www.epa.gov/OGWDW/methods/where.html.
2. 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.
3. M. Szelewski, "Fast Semivolatiles Analysis Using the Agilent Technologies 6890/5973 inert GC/MSD," Agilent Technologies, publication 5989-0207EN.
4. M. Szelewski, "Part-per-Trillion Calibration of Semivolatiles Using LVI-PTV-GC/MSD," Agilent Technologies, publication 5989-6589EN.
5. M. Szelewski, "Synchronous SIM/Scan Low-Level PAH Analysis Using the Agilent Technologies 6890/5975 inert GC/MSD," Agilent Technologies, publication 5989-4184EN.
6. H. Prest and J. Foote, "The Triple-Axis Detector: Attributes and Operating Advice," Agilent Technologies, publication 5989-7655EN.
7. R. Roushall and H. Prest, "The 5975C Series MSDs: Method Optimization and Trace Ion Detection," Agilent Technologies, publication 5989-6425EN.

For More Information

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

www.agilent.com/chem

Printed in the USA
April 14, 2008
5989-8342EN

Agilent Technologies