

Improved EPA-8330A Analysis with an Agilent InfinityLab Poroshell 120, 1.9 μm Column

Application Note

Environmental

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Abstract

A group of 14 nitroaromatics and nitramines was analyzed according to EPA Method 8330A for the trace analysis of explosive residues by HPLC/UV. The analysis was improved by using a superficially porous Agilent InfinityLab Poroshell 120, 1.9 μm column as compared to the traditional 5 μm column suggested by the United States Environmental Protection Agency (EPA). The 14 compounds were separated in 23 minutes on an InfinityLab Poroshell 120 EC-C18, 2.1 \times 150 mm, 1.9 μm column. Compared to the same analysis performed on an Agilent ZORBAX Eclipse Plus C18, 4.6 \times 250 mm, 5 μm column, the InfinityLab Poroshell column was able to run 3x faster while saving 88 % of the mobile phase and sample, and improving conditional peak capacity by 46 %. Both columns have similar selectivity, allowing easy method transferability.



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Introduction

Superficially porous particle LC columns are a popular tool in liquid chromatography. These columns generate high efficiency at lower pressure compared to their totally porous particle column counterparts [1]. This is primarily due to a shorter mass transfer distance and substantially narrower particle size distribution of the particles in the column [2]. The current trend with superficially porous particles is to reduce particle size for further efficiency improvements. The higher efficiency can be used to speed up analyses or improve results by increasing resolution and sensitivity.

Within the Agilent family of LC columns, significant work has been done to ensure easy method transferability and scalability. Stationary phase chemistries are very similar among totally porous particle Agilent ZORBAX and superficially porous particle Agilent InfinityLab Poroshell columns, so that methods can easily be updated without the need for further method development [3].

This work shows how small InfinityLab Poroshell particles can be used to improve the separation of 14 explosive residues found in EPA method 8330A compared to traditional 5 μm analyses. Significant time, solvent, sample, and money savings are demonstrated with the use of InfinityLab Poroshell columns.

Experimental

An Agilent 1290 Infinity LC System was used in this experiment. The system was modified from its standard configuration to have very low system volume and dispersion. Table 1 shows the configuration details, and lists the two Agilent LC columns that were used in this experiment.

Table 2 shows the LC method parameters for all analyses. Methods were geometrically scaled according to column volume to preserve the chromatographic separation from column to column.

Fourteen compounds were analyzed in this work; the EPA-8330A standard solution was purchased from Cerilliant. Methanol was purchased from Honeywell (Burdick and Jackson). Water was 0.2 μm filtered 18 MW from a Milli-Q system (Millipore).

Results and Discussion

The original separation of 14 explosive compounds is demonstrated on an Agilent ZORBAX Eclipse Plus C18, 4.6 \times 250 mm, 5 μm totally porous particle column. This separation was accomplished in 77 minutes, and is shown in Figure 1.

Table 1. UHPLC System Configuration

Agilent 1290 Infinity LC system configuration

Agilent 1290 Infinity Binary Pump (G4220A)	35 μL Solvent mixer: Jet weaver, 35 $\mu\text{L}/100 \mu\text{L}$ (G4220-60006)
Agilent 1290 Infinity High Performance Autosampler (G4226A)	Seat assembly, ultra low dispersion, for Agilent 1290 Infinity Autosampler (G4226-87030) Autosampler \rightarrow Heater: Capillary, stainless steel, 0.075 \times 220 mm, SV/SLV (5067-4784) Vial, screw top, amber with write-on spot, certified, 2 mL, 100/pk (5182-0716) Cap, screw, blue, PTFE/red silicone septa, 100/pk (5182-0717) Vial insert, 250 μL , glass with polymer feet, 100/pk (5181-1270)
Agilent 1290 Infinity Thermostatted Column Compartment (G1316C)	Heat exchanger, low dispersion, 1.6 μL , double (G1316-60005) Heater \rightarrow Column: A-Line quick-connect assembly, 105 mm, 0.075 mm (5067-5961) Column \rightarrow Flow cell: Capillary, stainless steel, 0.075 \times 220 mm, SV/SLV (5067-4784)
Agilent 1290 Infinity Diode Array Detector (G4212A)	Ultra-Low Dispersion Max-Light Cartridge Flow Cell, 10 mm (G4212-60038)
Agilent OpenLAB CDS ChemStation Edition Revision C.01.05 [35]	G4220A: B.06.53 [0013] G4226A: A.06.50 [003] G1316C: A.06.53 [002] G4212A: B.06.53 [0013]
Agilent LC columns	Agilent ZORBAX Eclipse Plus C18, 4.6 \times 250 mm, 5 μm (959990-902) Agilent InfinityLab Poroshell 120 EC-C18, 2.1 \times 150 mm, 1.9 μm (693675-902)

Table 2. UHPLC Method Parameters

Column	Mobile phase	Flow rate (mL/min)	Gradient	Injection volume (μL)	Sample	Thermostated column compartment (°C)	Diode array detector
Agilent ZORBAX Eclipse Plus C18, 4.6 × 250 mm, 5 μm	A) water B) methanol	1.0	25–35 %B in 77 minutes	8	Cerilliant ERE-021 Method 8330 Stock Standard 200 μg/mL each component in acetonitrile • 2-Amino-4,6-Dinitrotoluene • 4-Amino-2,6-Dinitrotoluene • 1,3-Dinitrobenzene • 2,4-Dinitrotoluene • 2,6-Dinitrotoluene • HMX • Nitrobenzene • 2-Nitrotoluene • 3-Nitrotoluene • 4-Nitrotoluene • RDX • Tetryl • 1,3,5-Trinitrobenzene • 2,4,6-Trinitrotoluene (TNT)	40	254 nm, 80 Hz
Agilent InfinityLab Poroshell 120 EC-C18, 2.1 × 150 mm, 1.9 μm		0.4	25–35 %B in 23 minutes	1			

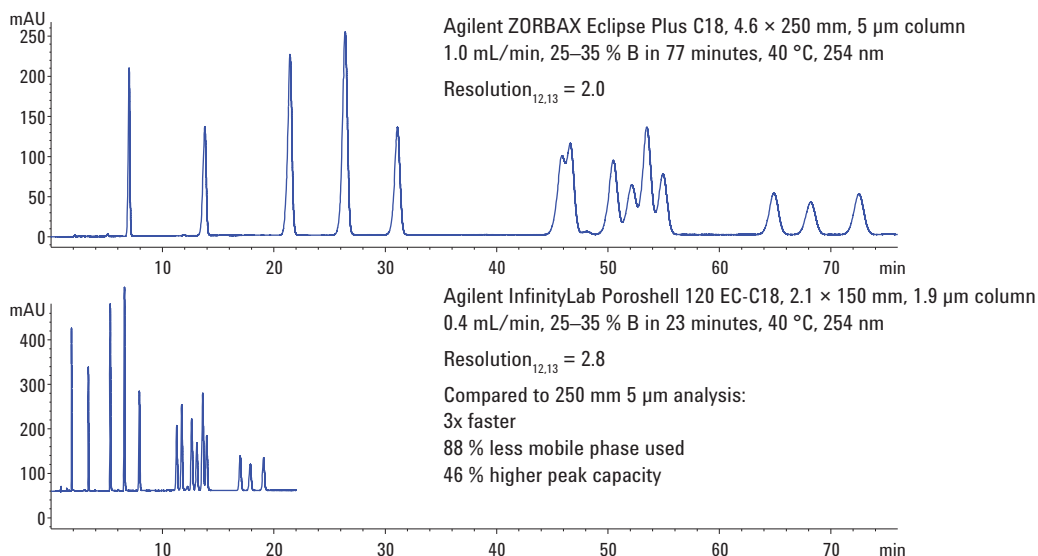


Figure 1. An Agilent ZORBAX Eclipse Plus C18, 4.6 × 250 mm, 5 μm analysis of explosive compounds found in EPA-8330 is improved by transferring to a high-performance Agilent InfinityLab Poroshell 120 EC-C18, 2.1 × 150 mm, 1.9 μm column; resolution and peak capacity are improved, while saving significant time, sample, solvent, and money.

Figure 1 also shows the same LC method transferred to a 2.1 × 150 mm, 1.9 μm superficially porous particle Agilent InfinityLab Poroshell 120 EC-C18. ZORBAX Eclipse Plus C18 and InfinityLab Poroshell 120 EC-C18 have very similar bonded chemistries for highly correlated overall selectivity, so no changes to the method were required. However, because the columns are different dimensions, the gradient, flow rate, and injection volume were geometrically

scaled for the smaller column volume. Flow rate was also increased for the InfinityLab Poroshell 1.9 μm column to run at the optimal flow rate for this smaller particle. The resulting chromatogram is 3x faster, uses 88 % less mobile phase and 88 % less sample, and improves conditional peak capacity by 46 %. Table 3 shows a summary and comparison of all chromatographic results.

Table 3. Comparison of EPA-8330 Analyses With Different LC Columns

Column	Pressure (bar)	Resolution ^{12,13}	Conditional peak capacity (nC)	Run time (min)	Mobile phase consumption (mL)				
Agilent ZORBAX Eclipse Plus C18, 4.6 × 250 mm, 5 µm	240	2.0	54	77	77				
Agilent InfinityLab Poroshell 120 EC-C18, 2.1 × 150 mm, 1.9 µm	1132	2.8	40 % Better resolution	79	46 % Higher peak capacity	23	3x Faster	9.2	Uses 88 % less mobile phase

Conclusions

The highly efficient Agilent InfinityLab Poroshell 120, 1.9 µm column can be used to improve existing methods using traditional columns, such as an Agilent ZORBAX Eclipse Plus C18, 5 µm column. The InfinityLab Poroshell and ZORBAX families offer similar bonded phase chemistries so that methods are easily transferred, often requiring no additional method development. When LC system pressure limits allow, the InfinityLab Poroshell 1.9 µm column can be used to accomplish faster separation, which will save significant time, solvent, sample, and money compared to traditional LC columns.

References

1. A. Gratzfield-Huguen, E. Naegele. Maximizing efficiency using Agilent Poroshell 120 Columns. *Agilent Technologies Application Note*, publication number 5990-5602EN, **2016**.
2. V. R. Meyer. Practical High Performance Liquid Chromatography. Fourth Edition, p. 34. Wiley (2004).
3. Transfer of Methods between Poroshell 120 EC-C18 and ZORBAX Eclipse Plus C18 Columns. *Agilent Technologies Application Note*, publication number 5990-6588EN, **2011**.

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