



"Better Surface Chemistry for Better Separation."

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Efficient Detection of Melamine Using Newly Developed HPLC Phases and Comprehensive Methods from Sepax

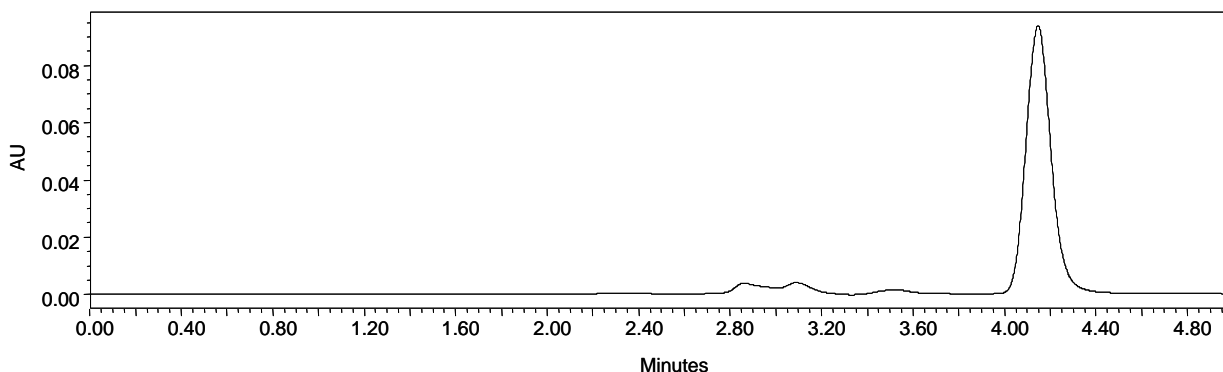
Even though ion-pair chromatographic methods have been traditionally used for melamine detection, its incompatibility with LC-MS limits its method from being widely used based on today's chromatographic detection sensitivity needs.

To further improve and better address needs of today's market, **Sepax Technologies, Inc. (Newark, DE)** has developed and launched series of NEW types of HPLC columns (HILIC Polar-100 and HP-SCX) ideal for LC-MS application and specifically catered toward efficient detection of melamine. For comparison purpose, chromatograms of Sepax C8 and C18 using ion-pair mobile phase are also included.¹

(1) HILIC Method

This method employs a HILIC Polar-100 column from Sepax Technologies, Inc (4.6 × 250mm, 5μm, P/N 131585-4625).

Column: HILIC Polar-100 (5μm, 120Å, 4.6 × 250m)
Mobile phase: 10mM NH₄Ac: acetonitrile = 10: 90 (v/v)
Flow rate: 1mL/min
Injection volume: 10μL
Wavelength: 240nm



Melamine was eluted at 4.15 min, tailing factor: 1.1; Plate#: 25000/m. This type of column can be used for rapid separation.

¹ For sample prep purpose, melamine was extracted using Sepax-UCT DBX (60mg/3mL, P/N SSDBX063). Details available from "Sample Preparation for Efficient Detection of Melamine" article.



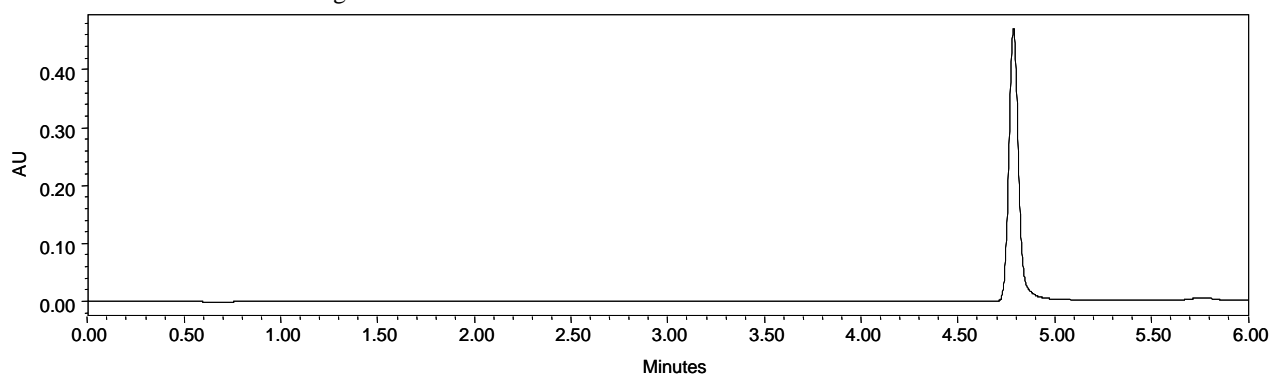
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(2) HP-SCX Method

This method employs a Sepax HP-SCX column (5 μ m, 120 \AA , 4.6 \times 250mm, P/N 120365-4625).

Column: Sepax HP-SCX (5 μ m, 120 \AA , 4.6 \times 250mm)
Mobile phase: 10mM NH₄Ac
Flow rate: 1mL/min
Injection volume: 10 μ L
Wavelength: 240nm



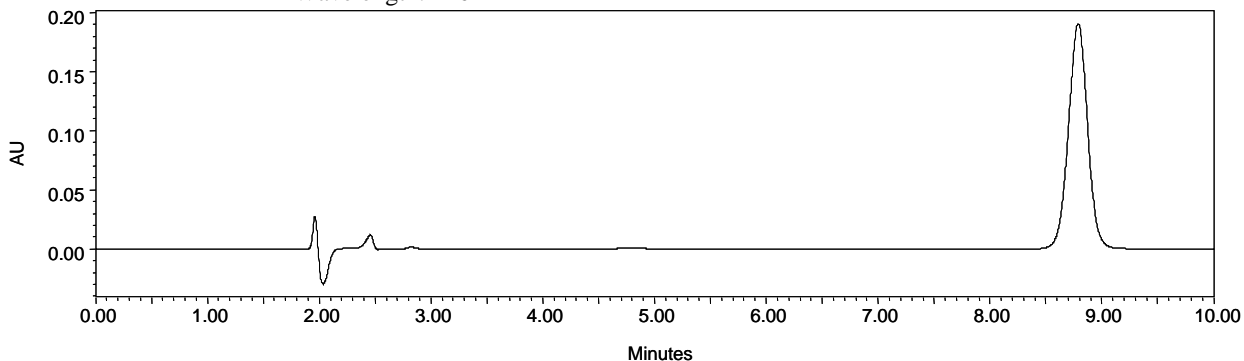
In this chromatogram, melamine eluted at 4.8 min, tailing factor: 1.2; Plate#: 180000/m. Such high efficiency and short retention time make HP-SCX the ideal column for rapid separation.

(3) Normal Methods

For traditional ion-pair chromatography, Sepax Technologies, Inc. offers Sepax GP-C8 (5 μ m, 120 \AA , 4.6 \times 150mm; P/N 107085-4615) and Sepax HP-C18 columns (5 μ m, 120 \AA , 4.6 \times 150mm; P/N 103185-4615).

a. Method I

Column: Sepax GP-C8 (5 μ m, 120 \AA , 4.6 \times 150mm)
Buffer: 10mM citric acid, 10mM sodium heptane sulfonate
Mobile phase: buffer /acetonitrile = 90/10 (v/v)
Flow rate: 1mL/min
Injection volume: 10 μ L
Wavelength: 240nm



Retention time of melamine: 8.8min, tailing factor: 1.0, plate #: 80000/m.

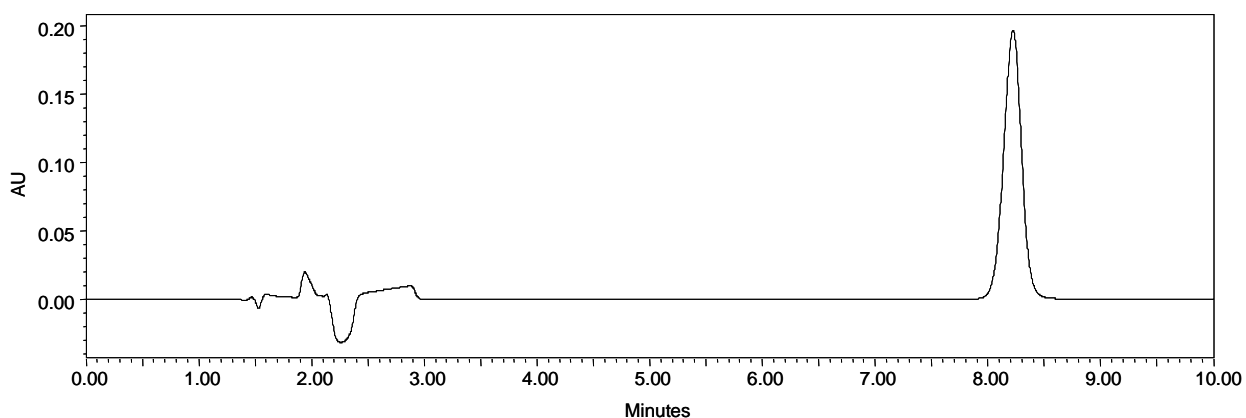


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b. Method II

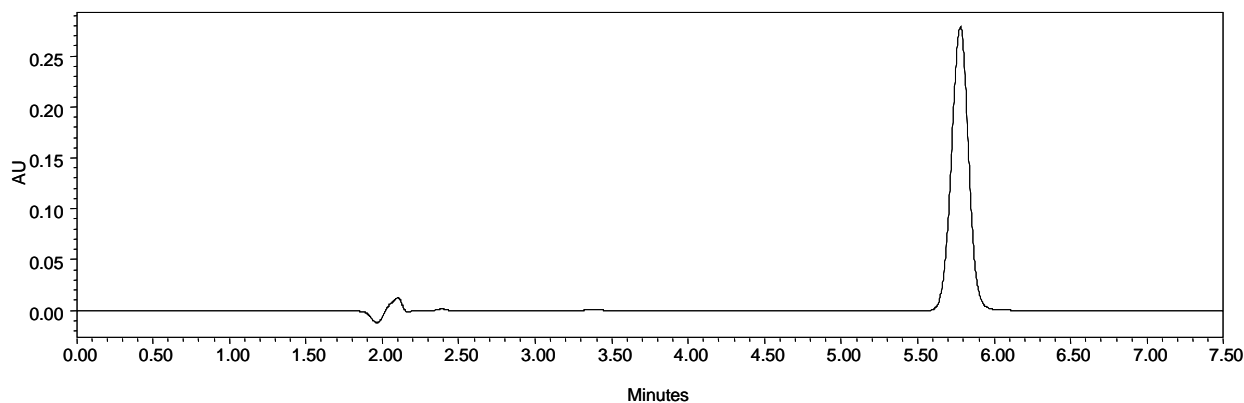
Column: Sepax GP-C8 (5 μ m, 120 \AA , 4.6x150mm)
Buffer: 10mM citric acid, 10mM sodium Octane sulfonate, pH: 3.0
Mobile phase: buffer/acetonitrile = 85: 15 (v/v)
Flow rate: 1.0mL/min
Injection volume: 10 μ L
Wavelength: 240nm



Retention time: 8.2min; tailing factor: 1.0; plate #: 80000/m.

c. Method III

Column: Sepax HP-C18 (5 μ m, 120 \AA , 4.6x150mm)
Buffer: 10mM citric acid, 10mM sodium heptane sulfonate
Mobile phase: buffer /acetonitrile = 90/10 (v/v)
Flow rate: 1mL/min
Injection volume: 10 μ L
Wavelength: 240nm



Retention time: 5.8min; tailing factor: 1.0; plate #:82000/m.