

APPLICATION NOTE

Mixed-Mode Stationary Phase compared with Polymeric Reversed Phase: Larger more retentive molecules

STYROS™ HQ-C8 versus STYROS™ 1R.

Mixed-Mode Chromatography or MMC also consists of using a hydrophobic alkyl chain on a quaternary amine anion.

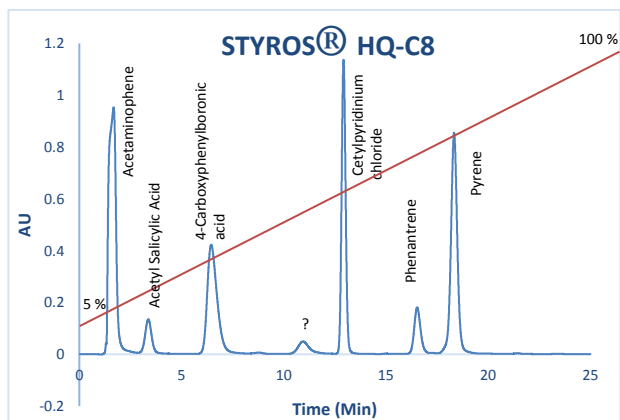
This mode can also be operated with a reversed phase regimen. In this Application Note we have chosen 6 compounds with strong hydrophobicity to test our reversed phase polymeric.

Pyrene and Phenanthrene as PAH and persistent pollutants are strongly adsorbed to the polymeric PS-DVB and fail to elute from it.

Even with an aggressive gradient of 50 to 100 % organic rich buffer.

The softer mixed mode phase, also polymeric, however milder hydrophobic tentacles, readily elutes and separates them with ample baseline separations.

The following chromatogram shows the separation on a 10 cm column.

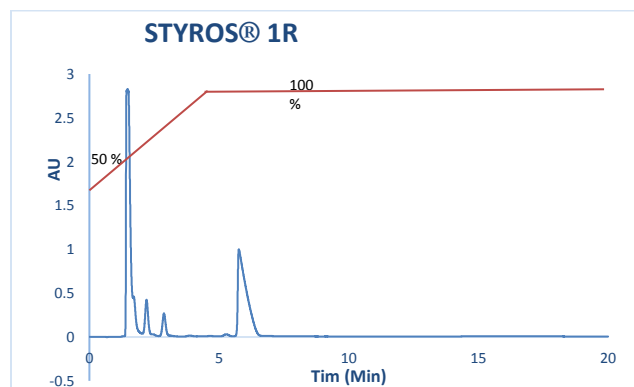


Chromatogram 1

Separation of 6 compounds on **STYROS® HQ-C8**

HPLC System.	Acquity UPLC / CLASS PLUS
Columns	STYROS® HQ-C8 Simulated-Monolith™ 2.1x100 mm
Mobile Phase For reversed phase.	A: 2% ACN in H2O with 0.075% TFA B: ACN: DI H2O (70:30) with 0.075% TFA
Flow rate	0.2 ml/min. Back pressure 1,337 psi (System included)
Gradient	5 to 100 % B in 25 minutes, 100 % B to 30 minutes.
Temperature	37°C
Detection	254 nm
Injection volume	6 µl
Sample:	6 compounds with various amounts from 1 to 5 mg/ml in buffer B.

The two columns are the same size, the gradient elution is markedly different to probe the possibility of the products to elute. The volumetric flow rate of 0.2 ml/min is similar.



Chromatogram 2

Injection of same 6 compounds on **STYROS® 1R**

HPLC System.	Acquity UPLC / CLASS PLUS
Columns	STYROS® 1R Simulated-Monolith™ 2.1x100 mm
Mobile Phase For reversed phase.	A: 2% ACN in H2O with 0.075% TFA B: ACN: DI H2O (70:30) with 0.075% TFA
Flow rate	0.2 ml/min. Back pressure 1,260 psi
Gradient	50 to 100 % B in 5 minutes, 100 % B to 20 minutes.
Temperature	37°C
Detection	254 nm
Injection volume	6 µl
Sample:	6 compounds with various amounts from 1 to 5 mg/ml in buffer B.

In the previous Application Note using smaller compounds the difference between the two phases was displayed in the longer retention in the reversed phase polymeric of the molecules tested.

This application Note highlights the higher limits of the MMC and its usefulness in use as multimodal chromatography with broader use.

The Mixed Mode or MMC operates on a hydrophilic base of quaternary amine to which a hydrophobic tail operating on the van der Waals forces is attached.

Whereas the PS-DVB operates solely on aryl entities and π - π interactions.

