

APPLICATION NOTE

Quaternary Amino Methyl (HQ) Anion Exchanger: Beyond Mono Q

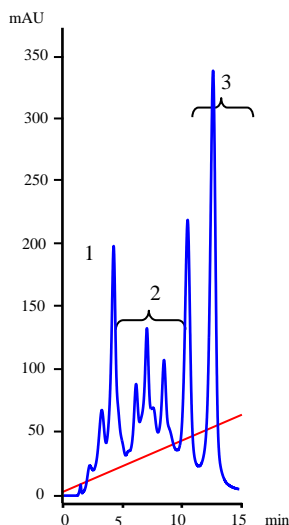
Anion exchanger chromatography with quaternary amine functions is a common mode of separation using media with permanently charged surface.

It is also called SAX or Strong Anion Exchanger chromatography indicating the constancy of charges independent of the pH.

In Application Note 59 **STYROSTM HQ Simulated MonolithTM** was compared with Mono Q HR 16/10 using its function test under similar conditions.

Unlike monolith, Simulated MonolithTM columns have high dynamic capacity and high resolution in addition to low back pressures.

A separation that takes 30 minutes on a Mono Q column can be achieved in half the time by taking advantage of the higher efficiency of **STYROSTM Simulated MonolithTM** media.



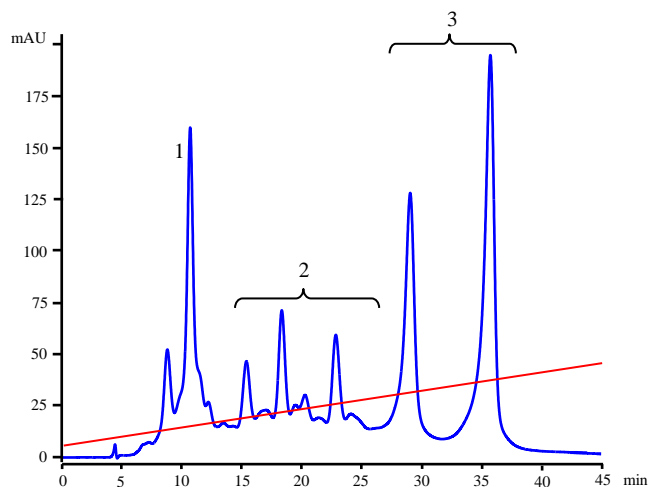
STYROSTM HQ Simulated MonolithTM 4.6 x 100 mm Stainless Steel.

Table 1. Operating parameters.

| | |
|-------------------------|---|
| HPLC System. | Agilent 1100 with thermostatted column compartment. |
| Columns | STYROSTM HQ 4.6 X 100 mm Stainless Steel. 1.66 ml, column volume |
| Mobile phase. | A: 20 mM Piperazine, pH=6 B: A + 0.3 M NaCl, pH=6 |
| Flow rate | 0.8 ml/min (290 cm/hr of linear flow rate) |
| Gradient | 0.6 to 100 % B in 15 min (7 cv) |
| Temperature | 30°C |
| Detection | 280 nm |
| Injection volume | 100 µl |
| Sample: | 1. Transferrin (human) 2 mg/ml, 2. Ovalbumin, 4 mg/ml, 3. b-Lactoglobulin, 4 mg/ml |

The back pressure of the column is 6 bar (87 psi) at 0.8 ml/min. and 30 °C.

Under similar conditions, the back pressure of a **4.6 x 300 mm** column increases only to 16 bar (232 psi) providing higher resolution for the separation.



STYROSTM HQ Simulated MonolithTM 4.6 x 300 mm Stainless Steel.

Table 2. Operating parameters.

| | |
|-------------------------|---|
| HPLC System. | Agilent 1100 with thermostatted column compartment. |
| Columns | STYROSTM HQ 4.6 X 300 mm Stainless Steel. 4.98 ml, column volume |
| Mobile phase. | A: 20 mM Piperazine, pH=6 B: A + 0.3 M NaCl, pH=6 |
| Flow rate | 0.8 ml/min (290 cm/hr of linear flow rate) |
| Gradient | 0.6 to 100 % B in 45 min (7 cv) |
| Temperature | 30°C |
| Detection | 280 nm |
| Injection volume | 100 µl |
| Sample: | 1. Transferrin (human) 2 mg/ml, 2. Ovalbumin, 4 mg/ml, 3. b-Lactoglobulin, 4 mg/ml |

High dynamic capacity, high resolution and low back pressure of **STYROSTM HQ Simulated MonolithTM** provide the appropriate columns for Simulated Moving Bed chromatography.

SMB is necessary in addressing the limitations of downstream processing as it presently stands.

Simulated Moving Bed chromatography can reduce chromatography media volume and buffer consumption per unit of productivity.

The use of multiple channels provides high cumulative flow rates.

While the use of monolith in Simulated Moving Bed chromatography is an improvement over the use of conventional media, **Simulated MonolithTM** is the next step in this process as it addresses the capacity limitations of monolith while reducing the back pressures and preserving the resolution of high performance media.