

APPLICATION NOTE

Comparison of **STYROS™ 1R/XP** and **1R/XH**.

The **STYROS™** chromatographic stationary phase family of products has been developed to permit a seamless transition from analytical to preparative scale.

In the past, researchers had to make several compromises concerning pressure drop and efficiency.

Small particle sizes were used for analytical separations, in order to maximize the theoretical plates. This resulted in considerable back pressure.

Due to the hardware limitations and cost considerations, large-size particles have been used in micro preparative and preparative scale applications, resulting in low back pressure at the expense of significant loss in theoretical plates as compared with analytical columns.

We have recognized at **OraChrom**, the need for a chromatographic media that provides both the high performance and the low back pressure at the same time. Due to our carefully optimized polymerization technique, the special particle size distribution and the high pressure tolerance of **STYROS™** media, it is now possible to provide packed columns for high performance analytical as well as preparative applications without necessarily sacrificing the efficiency.

The **STYROS™ XP** series have been developed to provide high resolution separations with minimal back pressure.

The XH series, with over 35,000 plates/m, are intended more as analytical columns for fast separations and optimal back pressure.

The N/ΔP ratio (N: theoretical plates, ΔP: pressure drop across the column) is an important factor that has great significance in determining the practicality of an LC column as a component of a large scale preparative process.

The chromatographic conditions are gathered in the following table:

Table 1. Operating Parameters For Chromatograms 1-4.

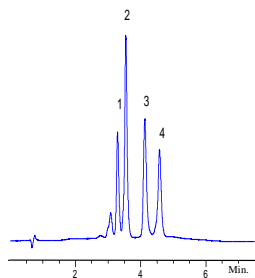
HPLC System	Hewlett Packard 1050
Detector	214 nm
Column	STYROS™ 1 R/XH 50x4.6 mm (Chromatograms 1 & 2) STYROS™ 1 R/XP 50x4.6 mm (Figure 3 & 4)
Mobile Phase	A: 0.1 % TFA in water B: 0.1 % TFA in Acetonitrile/water 95/5
Gradient	15-80% B in 10 ml
Flow rate	1 and 5 ml/min
Temperature	Ambient
Injection volume	10 µl
Sample 1 mg/ml each	1: Cytochrome C, 2: Lysozyme 3: β-Lactoglobulin, 4: Ovalbumin

The XP series of **STYROS™** shows an N/ΔP ratio of $\cong 900$ for a 50x4.6 mm column. A typical reversed-phase column packed with 5 µm silica particles with 60,000 plates/m, has a ratio of $\cong 200$.

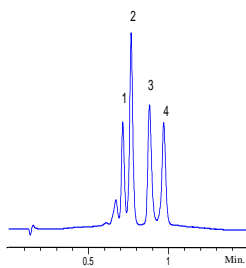
The following chromatograms highlight the performance of the XH and XP series and compare them with one another at different flow rates.

The XP series provide a more accurate picture of what the resolution would be at larger scale without having to actually use large amounts of sample and high volume of solvent in order to determine the chromatographic conditions.

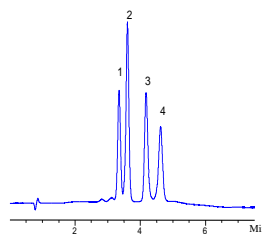
Another alternative is the M series of **STYROS™** that offer closer efficiency number to the preparative columns providing closer similitude during the transition to preparative scale.



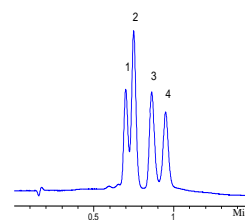
Chromatogram 1.
Standard Protein Separation on
STYROS™ 1 R/XH at 1 ml/min
(360 cm/hr).



Chromatogram 2.
Standard Protein Separation on
STYROS™ 1 R/XH at 5 ml/min
(1,800 cm/hr).



Chromatogram 3:
Standard Protein Separation on
STYROS™ 1 R/XP at 1 ml/min
(360 cm/hr).



Chromatogram 4.
Standard Protein Separation on
STYROS™ 1 R/XP at 5 ml/min
(1,800 cm/hr).