High throughput and robust SEC measurement using newly developed all-in-one SEC instrument for polymers

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Introduction

- The EcoSEC® GPC system is an innovative, all-in-one instrument that results in unprecedented reproducibility due to advanced flow and temperature control. The instrument consists of an in-line degasser, auto-sampler, temperature-controlled pumping system, column oven, refractive-index (RI) detector, and optional UV detector. All components are housed in the chassis and controlled via the EcoSEC Workstation software.

- The fundamental characteristics of this new GPC/SEC system were assessed and the results are reported. We investigated the precision and the reproducibility of flow rate, the precision of temperature control in the pumping unit, column oven and RI detector, the base line stability against external temperature fluctuations, the sensitivity of the RI detector, and other system features. Furthermore, the precision and the reproducibility of molecular mass and molecular mass distribution were examined using semi-micro TSK-GEL SuperMultiporeHZ Series columns and PStQuick polystyrene calibration markers.

Experimental Conditions

Chemicals

- Stabilized analytical grade THF used without further treatment (Wako, Japan).
- Synthetic polymers and other reference standards (Wako and other resin suppliers).
- Polystyrene standards with narrow molecular-mass distribution (Tosoh).

Columns (Tosoh)

- TSKgel® SuperMultiporeHZ-H, 4.6mm ID x 15cm, 6µm
- TSKgel SuperMultiporeHZ-M, 4.6mm ID x 15cm, 4µm
- TSKgel SuperMultiporeHZ-N, 4.6mm ID x 15cm, 3µm
- TSKgel MultiporeHxl-M, 7.8mm ID x 30cm, 5µm
- TSK-GEL® SuperHZ type columns, 4.6mm ID x 15cm

Other Columns

- Mixed-bed type columns (4.6mm ID x 25cm) were obtained from Varian, Inc.

Preparation of sample solutions

- Synthetic polymers were dissolved in THF at concentrations of 0.1-0.0125g/L.
- Polystyrene standards were dissolved in THF at concentrations of 0.2-1.0g/L and gently stirred for a period of 12-24 hours prior to use.

Results and Discussion

- **Figure 1.** The EcoSEC GPC System is equipped with an in-line degasser, pumping unit, auto-injector, column oven, and RI detector. The pumping system, tubing and RI detector are enclosed in a temperature-controlled environment

- **Figure 2.** Ten microliters of polystyrene samples (M_w/M_n= 1.01) were injected onto a TSKgel SuperHZ-M column using THF as a solvent. Temperature was cycled 23°C to 26°C.
  - ***The retention volume (mL), peak maximum %CV for the EcoSEC GPC System was 0.07 (n=10) versus a %CV of 0.23 (n=10) for a conventional GPC system.***
  - ***Controlling the temperature of the pump housing, columns and tubing improves the baseline stability by removing the effect of temperature fluctuations.***

- **Figure 3.** Ten injections per day were run using two narrow MW samples, F-10 (polystyrene standard, Mw = 100,000; Mw/Mn=1.01) and F-2 (polystyrene standard, Mw =17,000; Mw/Mn=1.01). Each sample was injected separately.
  - ***With the exception of day five, the %CV was measured at < 0.03.***
  - ***The advanced pump design employs advanced check valve engineering and fluidics, which produces a consistent flow rate, resulting in high reproducible retention times.***

Results and Discussion

- **Figure 4.** Ten injections per day were run using vinyl chloride/vinyl acetate/vinyl alcohol terpolymer M_W ~30,000 Da.
  - ***The molecular weight was calculated and the %CV was found to be < 0.2% within each day.***
  - ***The instrument is engineered to produce less flow rate variations and as a consequence, a higher precision in molecular mass determinations is obtained.***

- **Figures 5a & 5b.** The EcoSEC GPC system features a dual flow RI detector. Thus any minor changes in solvent composition are "zeroed" out by the detector and therefore the baseline remains stable. In contrast, in a single flow design, the properties of the solvent in the reference cell slowly alter over time.
  - **With the EcoSEC GPC system, superposition of five consecutive chromatograms demonstrates negligible baseline drift (Figure 5a) as compared to the same experiment repeated with a stagnant reference cell (Figure 5b) from a conventional GPC unit.**

- **Figure 6.** The instrument is designed to minimize system dead volume.
  - **A low dead volume results in less extra-column band broadening and allows the use of shorter and narrower ID columns resulting in shorter run times and sharper peaks.**

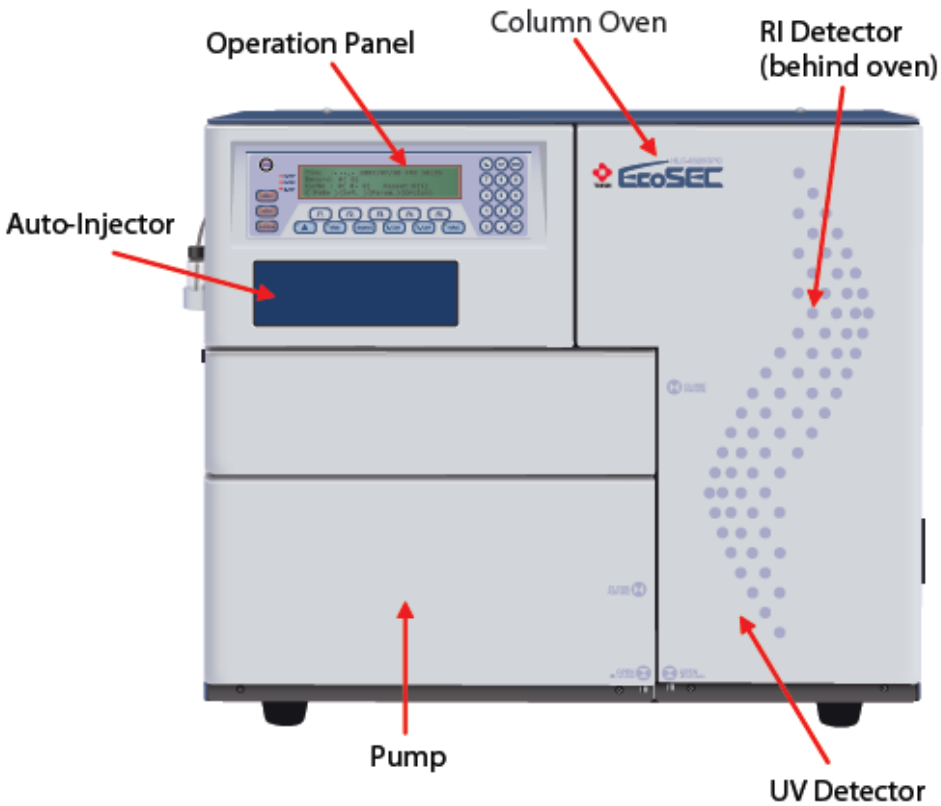
Results and Discussion

- **Figure 7.** The semi-micro layout of the system permits the use of smaller GPC columns.
  - **When using the TSK-GEL SuperMultiporeHZ Series of columns, analysis times are lowered by ½ compared to conventional columns and systems, while reducing the solvent consumption by 1/6th.**
- **Figure 8.** Consistent results are obtained on different EcoSEC GPC instruments run by different operators.
- **Table 1.** Changes in elution times significantly impact SEC analysis.
  - **The advanced engineering of the EcoSEC GPC system yields a high precision in retention time by reducing variation among instruments even when run by different users.**
- **Figure 9.** To take advantage of the semi-micro features of the EcoSEC GPC System, Tosoh introduced a new line of GPC columns, the TSK-GEL SuperMultiporeHZ Series of semi micro-columns (4.6mm ID). Unlike other columns that utilize a mixed resin bed, Tosoh has developed particles with multiple pore sizes on the particle using proprietary multi-pore technology.
- **Figure 10.** The multi-pore packing has a broad range of molecular weight fractions because each particle has a wide pore distribution.

Results and Discussion

- **Figure 11.** The TSK-GEL SuperMultiporeHZ columns are packed with monodisperse particles of uniform diameter, resulting in a high theoretical plate number.
- **Table 2.** Physical properties of TSK-GEL SuperMultiporeHZ columns.
- **Figure 12.** The calibration curves of the TSK-GEL SuperMultiporeHZ columns have excellent linearity, thus accurate analysis of molecular weight.
- **Figures 13a & 13b.** Chromatograms of various polymers analyzed using a TSKgel SuperMultiporeHZ-H column. Smooth chromatograms without any distortion are obtained for each of these polymers using different TSK-GEL SuperMultiporeHZ columns.

EcoSEC GPC System Features



Specifications

1. Pump

Flow Rate:	10 to 2,000µL/min
Accuracy:	+/- 2%
Precision:	+/- 0.2%
Max pressure:	25MPa

2. Auto-Injector

Injection vol.:	1 to 1,500µL
# of samples:	100

3. Column oven

Temp range:	ambient +10 to 60°C
Capacity:	eight, 7.8mm ID x 30cm columns

4. Detector(RI)

Type:	Dual flow type
Cell Volume:	2.5µL

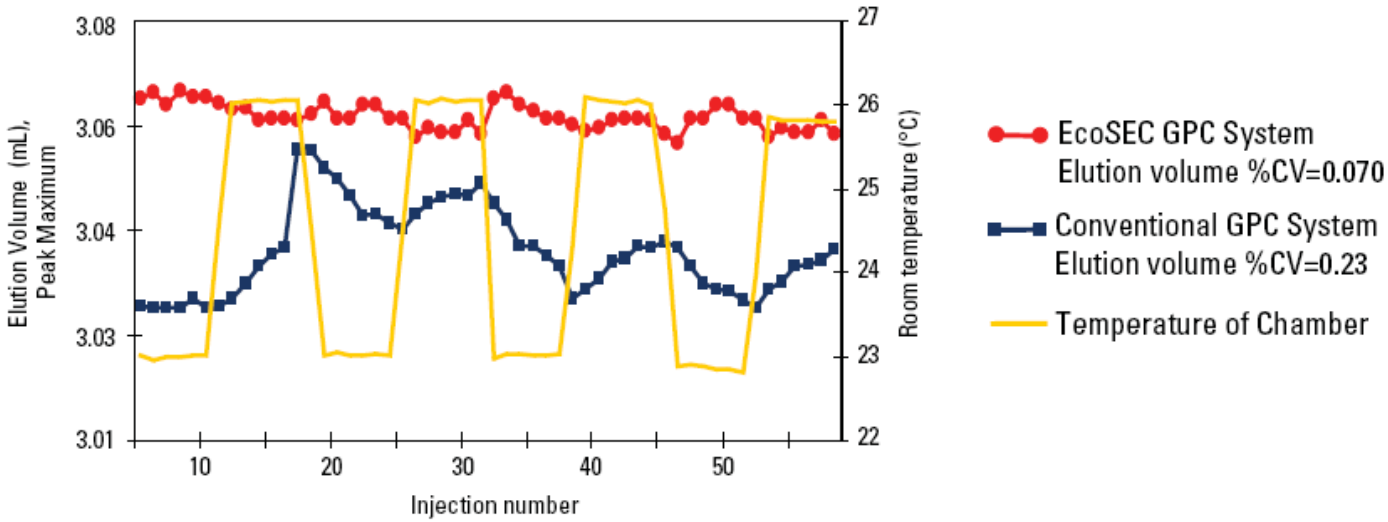
5. Detector(UV): optional

Wave length:	195 to 350nm
Cell volume:	2µL

6. Dimensions

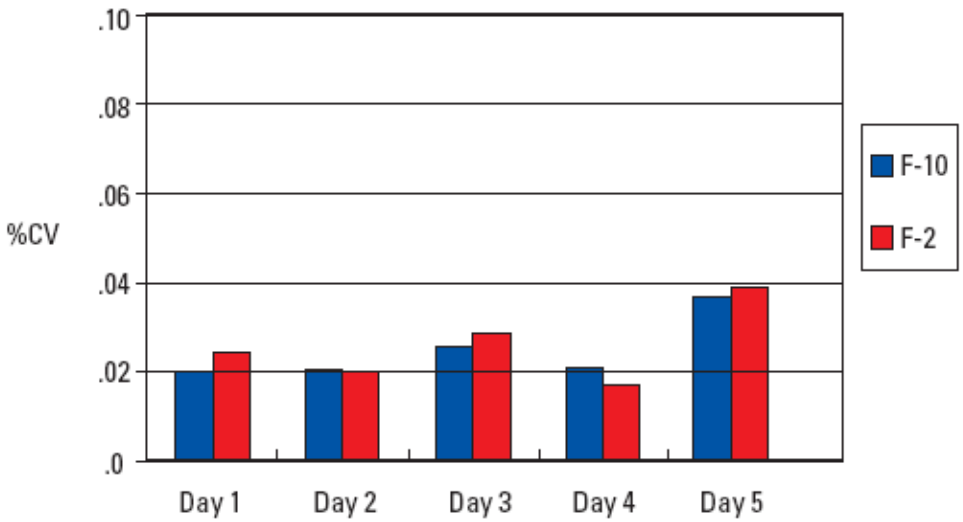
680(W) x 500(D) x 580(H)mm

Solvent Delivery Reproducibility with Changes in Room Temperature



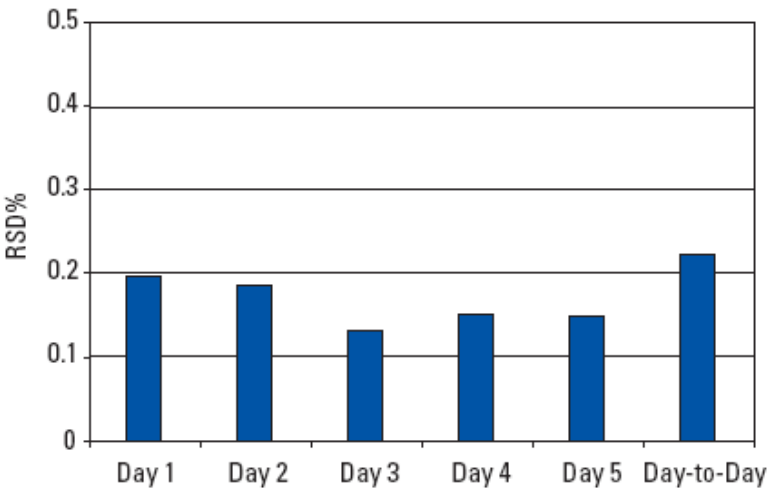
Column:	TSKgel SuperMultiporeHZ-M, 4µm, 4.6mm ID x 15cm x 2
Eluent:	THF
Detector:	RI
Flow Rate:	0.35mL/min
Sample:	Std polystyrene (Mw: 17,000)

Reproducibility of Retention Time



TSKgel SuperMultiporeHZ-M, 4µm, 4.6mm ID x 15cm x 2
THF
0.35mL/min
10/day
F-10, standard polystyrene (MW ~100,000; Mw/Mn=1.01)
F-2, standard polystyrene (MW=17,000; Mw/Mn=1.01)

Molecular Weight (M_W) Reproducibility

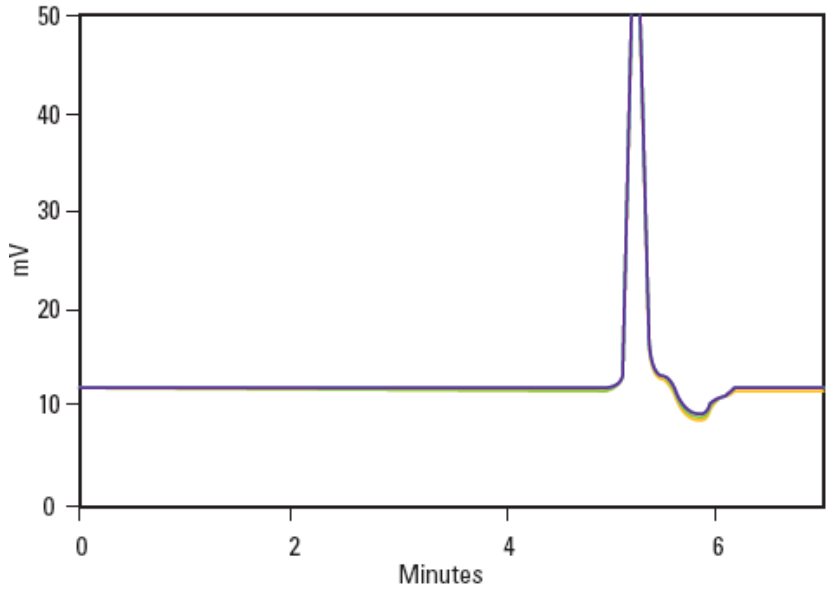


Column:	TSKgel SuperHM, 4µm, 4.6mm ID x 15cm x 2
Eluent:	THF
Flow Rate:	0.35mL/min
Number of injection:	10/day
Samples	vinylchloride/vinylacetate/ vinylalcohol terpolymer (2g/L, 20µL) (broad MWD polymer, Mw ~ 10,000Da)

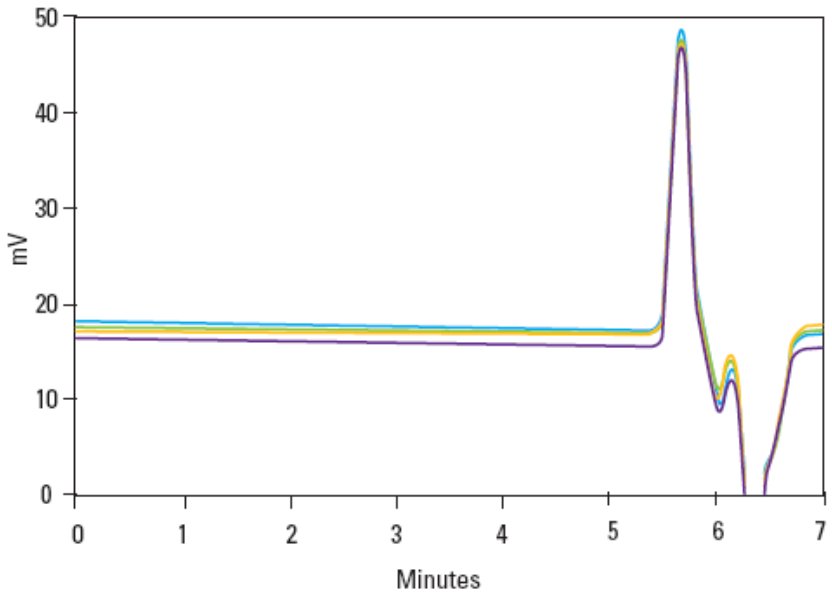
RSD%
- < 0.18% within day
- < 0.22% day-to-day

RI Baseline Match: Dedicated versus Component GPC System

Dual Flow RI Detector in EcoSEC GPC System

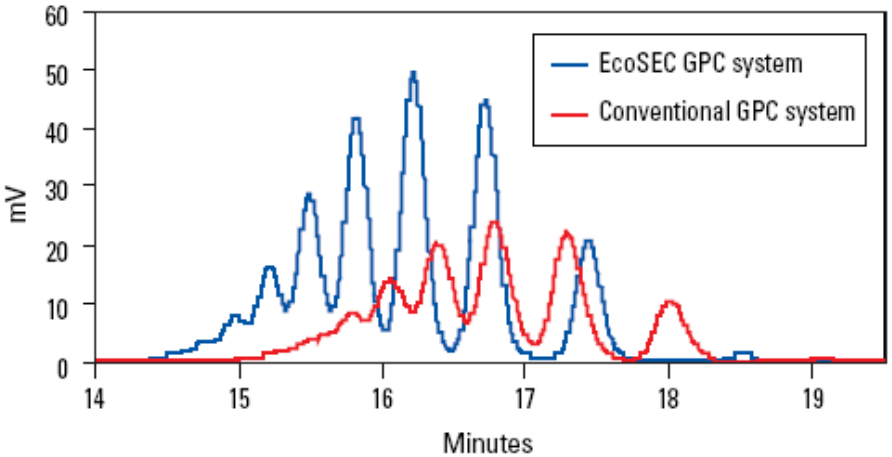


Single Flow RI Detector from Competitor GPC Instrument



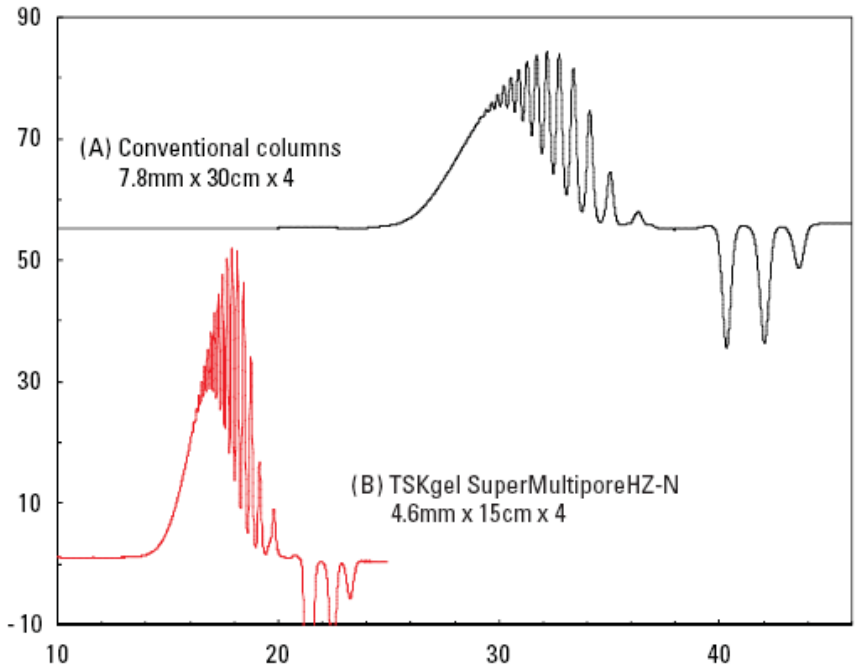
Column:	TSKgel SuperHZ-H, 6µm, 4.6mm x 15cm
Eluent:	THF
Detector:	RI
Temp.:	40°C
Flow rate:	0.35mL/min,
Sample:	dicyclohexylphthalate
Number of Injections:	5

Comparing Semi-micro and Conventional GPC Systems



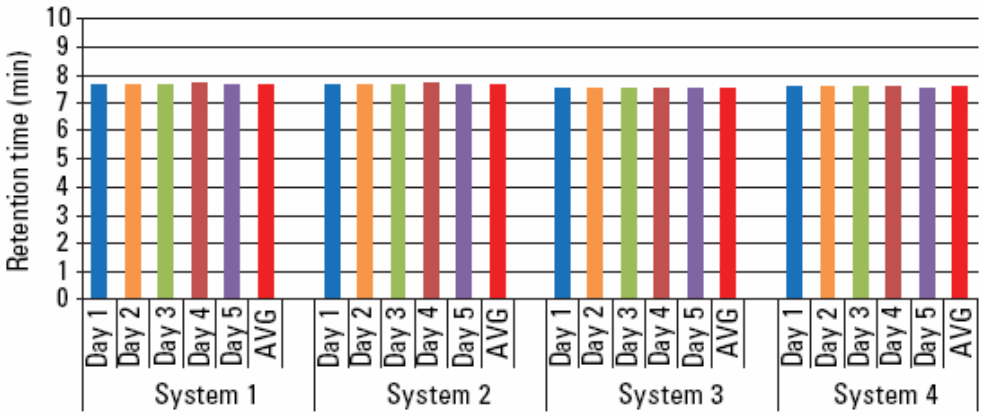
Column:	TSKgel SuperHZ2000, 6µm, 4.6mm ID x 15cm x 4
Eluent:	THF
Flow Rate:	0.35mL/min
Detector:	RI
Temp.:	40°C
Sample:	polystyrene oligomer (Mw~500)
Inj. volume:	10µL (0.2g/L)

Comparing Semi-micro and Conventional Columns



Eluent:	THF
Flow Rate:	(A) 1.0mL/min, (B) 0.35mL/min
Detector:	RI
Temp.:	40°C
Sample:	PTMEG, poly(tetramethyleneether glycol)
Inj. volume:	(A) 60µL (B) 10µL

Day-to-Day Reproducibility of Retention Time



Four Column:	TSKgel SuperMultiporeHZ-M, 4µm, 4.6mm x 15cm x 2
Eluent:	THF
Flow rate:	0.35mL/min
Detector:	RI
Temp.:	40°C
Sample:	vinylchloride/vinylacetatecopolymer 0.2% (broad MWD polymer, Mw ~ 10,000Da)
Inj. volume:	10µL

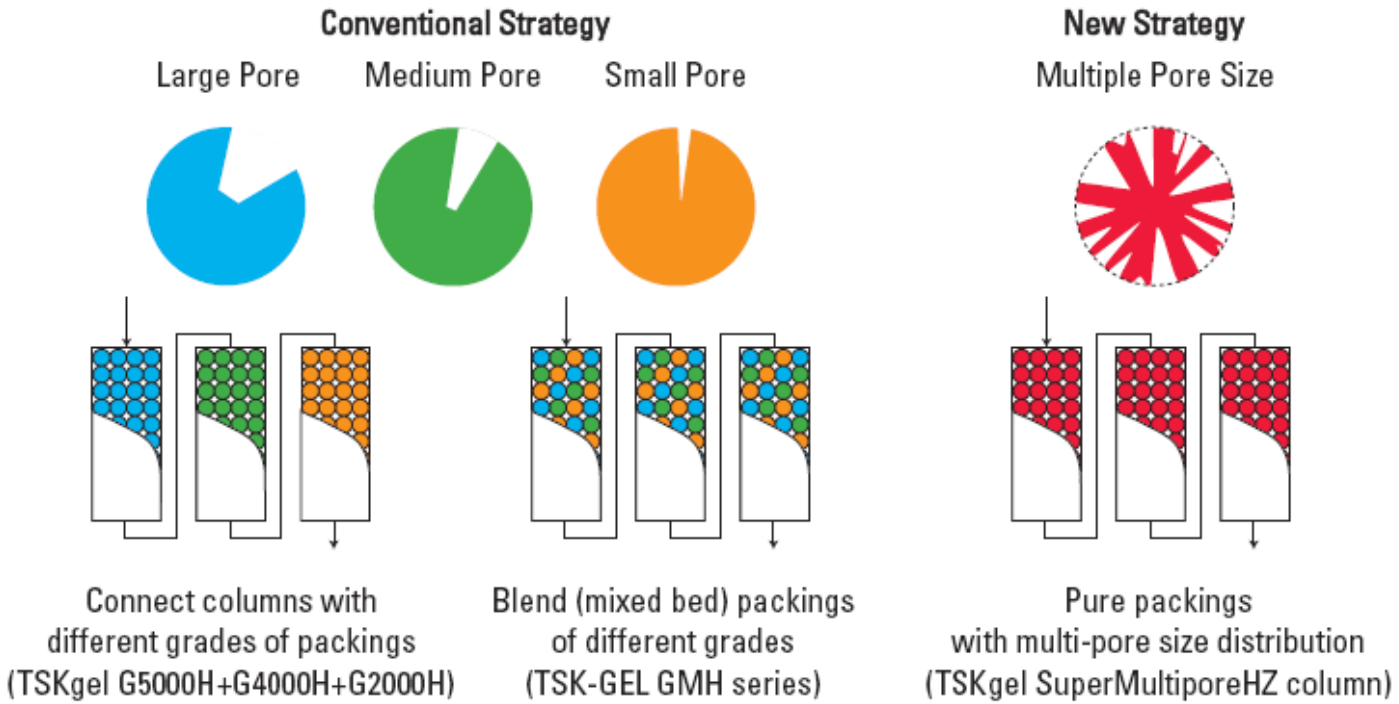
Four EcoSEC GPC systems, 4 operators, 4 column sets, 4 conditions, one location

System-to-System Reproducibility

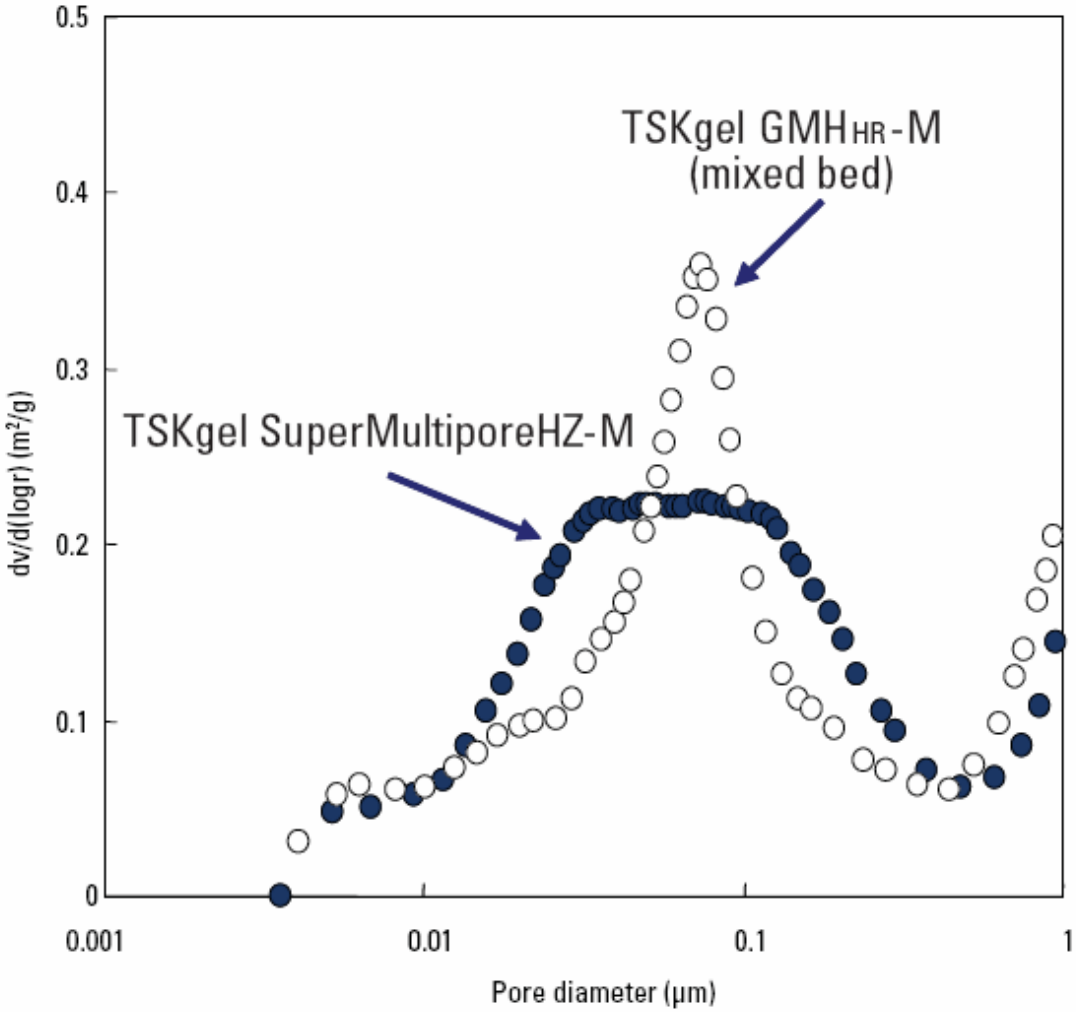
EcoSEC GPC System	M_n	%CV	M_w	%CV
A	13,802	0.496	29,770	0.157
B	13,747	1.483	29,821	0.193
C	13,641	1.448	29,814	0.175
D	13,632	1.152	30,041	0.113
Average	13,706	0.605	29,862	0.408

Four EcoSEC GPC systems, 4 operators, 4 column sets, 4 conditions, one location

Proprietary Multi-pore Particle Synthesis Technology

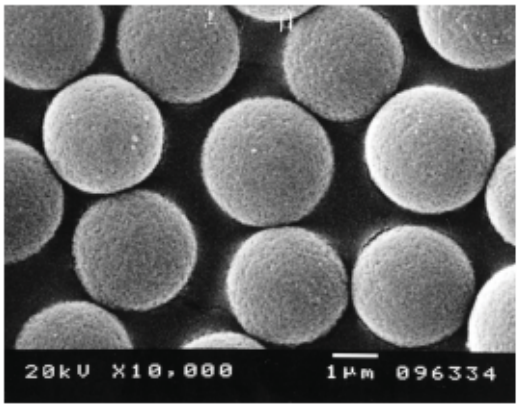


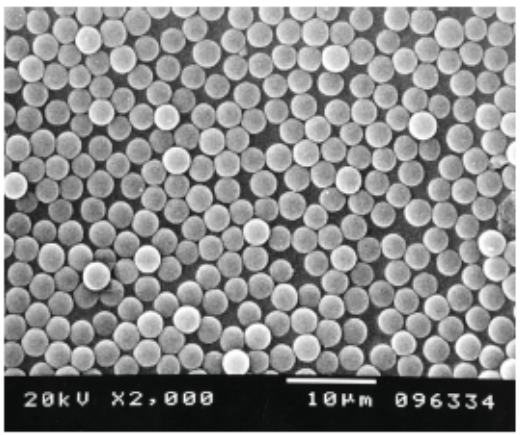
Pore Characteristics of TSK-GEL SuperMultiporeHZ-M and Mixed Bed Type Particles



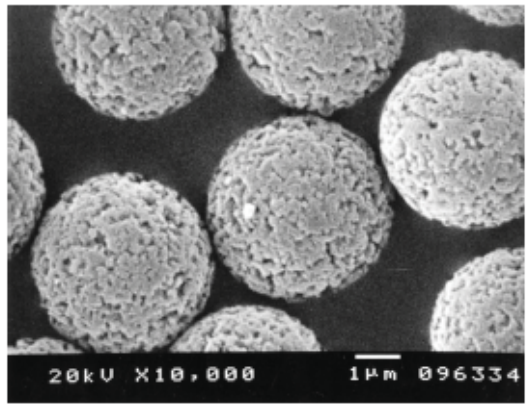
SEM Pictures of Multi-pore Particles

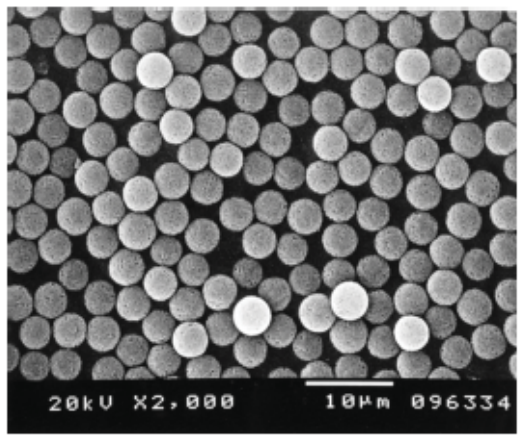
TSKgel SuperMultiporeHZ-N (3µm)



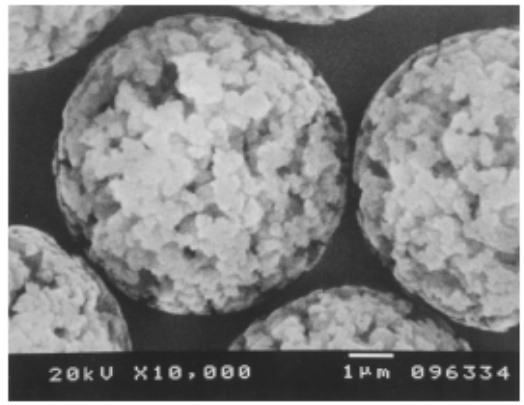


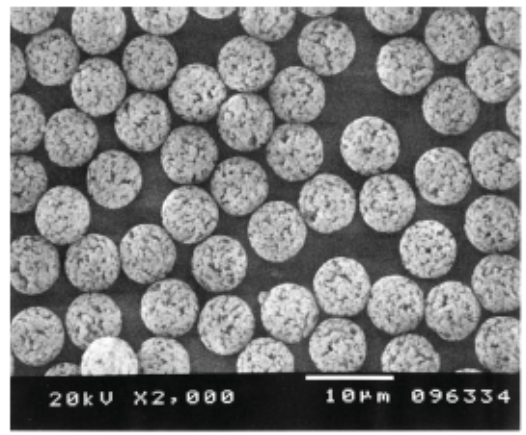
TSKgel SuperMultiporeHZ-M (4µm)





TSKgel SuperHZM-M (mixed-bed) (6µm)



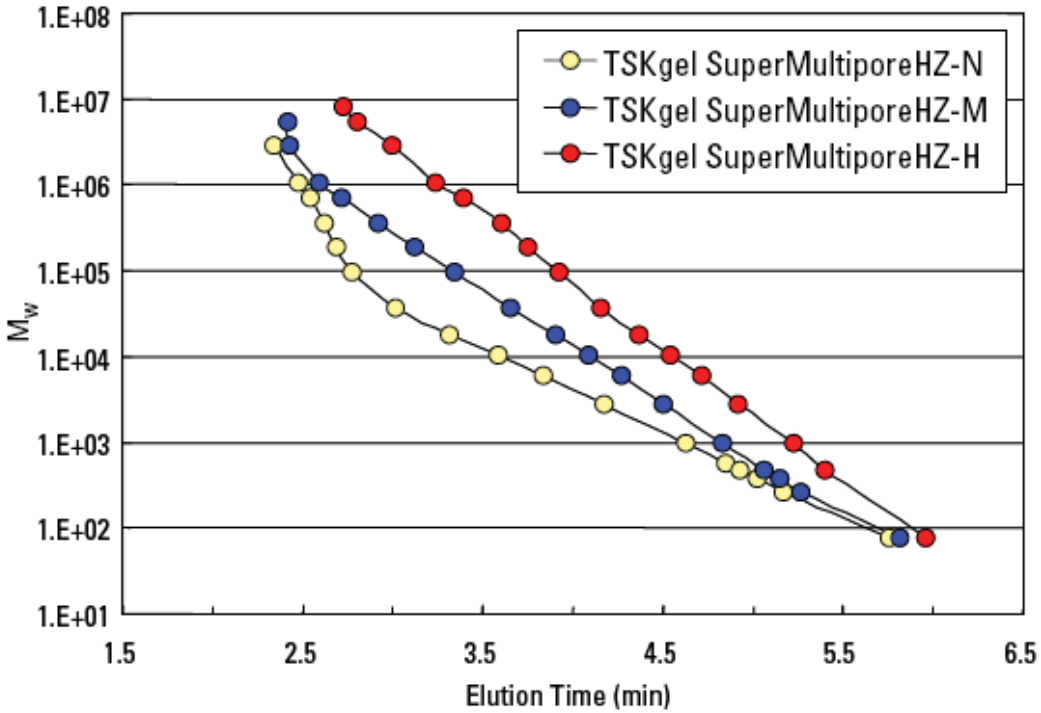


Physical Properties of TSK-GEL SuperMultiporeHZ Columns

Product Name	TSK-GEL SuperMultiporeHZ-N	TSK-GEL SuperMultiporeHZ-M	TSK-GEL SuperMultiporeHZ-H
Base material	Poly(styrene/divinylbenzene)	Poly(styrene/divinylbenzene)	Poly(styrene/divinylbenzene)
Particle size	3µm	4µm	6µm
Max. exclusion limit MW	120,000	2,000,000	40,000,000*
Mean pore size	80Å	140Å	--------
MW separation range	300-50,000	500-1,000,000	1,000-10,000,000
Theoretical plates/column	20,000	16,000	11,000
Column size (Analytical)	4.6mm ID x 15cm	4.6mm ID x 15cm	4.6mm ID x 15cm

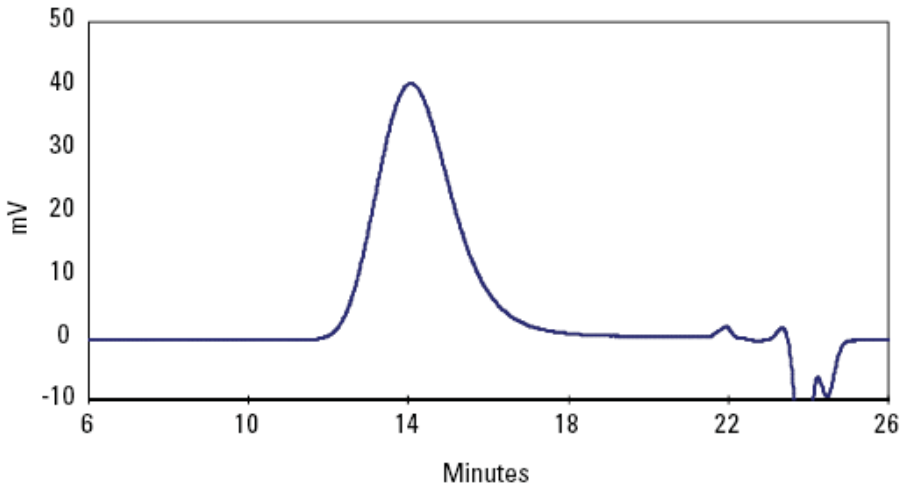
*Polystyrenes in THF

Calibration Curves of TSK-GEL SuperMultiporeHZ Columns



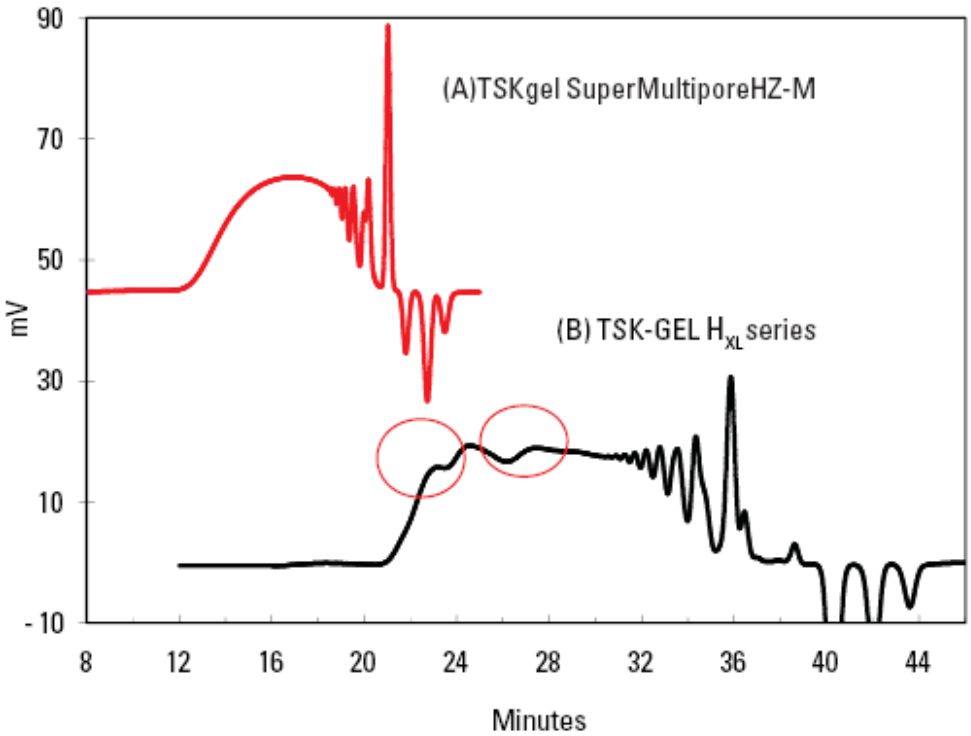
Columns:	TSKgel SuperMultiporeHZ-N, 3µm, 4.6mm ID x15cm TSKgel SuperMultiporeHZ-M, 4µm, 4.6mm ID x15cm TSKgel SuperMultiporeHZ-H, 6µm, 4.6mm ID x15cm
Eluent:	THF
Flow rate:	0.35mL/min
Temp:	25°C
Detection:	UV@254nm (UV-8020 w/ 2.4µL cell)
Sample:	PStQuick polystyrene standards

Analysis of Polyisobutylene on TSK-GEL SuperMultiporeHZ-M Columns



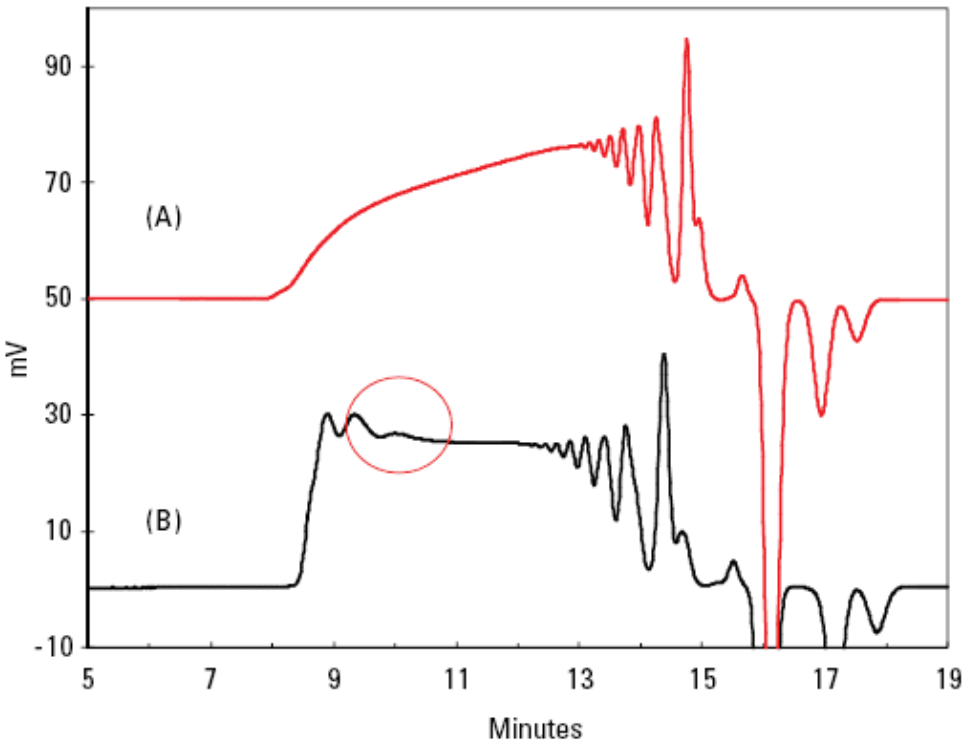
Column:	SuperMultiporeHZ-M, 4µm, 4.6mm x 15cm x 4
Eluent:	THF
Flow rate:	0.35mL/min
Temp.:	40°C
Detection:	RI
Sample:	Polyisobutylene (3µg/mL)
Inj. volume:	10µL

Chromatograms of Phenol Resin Obtained with TSK-GEL SuperMultiporeHZ-M Columns and Mixed Bed Columns From a Competitor



Columns:	(A) TSKgel SuperMultiporeHZ-M, 4µm, 4.6mm ID x 15cm x 4 (B) TSKgel G4000HXL+3000+2500+2000 7.8mm ID x 30cm x 4
Solvent:	THF
Flow rate:	(A) 0.35mL/min (B) 1.0mL/min
Detection:	RI
Temp.:	40°C
Sample:	Phenol resin (3µg/µL)
Inj. volume:	(A) 10µL (B) 50µL

Chromatograms of Phenol Resins Created with the TSK-GEL SuperMultiporeHZ-N Columns and TSK-GEL SuperHZ Series Columns



Column:	(A) TSKgel SuperMultiporeHZ-N, 3µm, 4.6mm ID x 15cm x 3 (B) TSKgel SuperHZ 3000+2500+2000, 4.6mm ID x 15cm x 3
Solvent:	THF
Flow rate:	0.35mL/min
Detection:	RI
Temp.:	40°C
Sample:	Phenol resin (3µg/µL)
Inj. volume:	10µL

Conclusions

- The results demonstrate that the new EcoSEC GPC system is suitable for high-throughput SEC as well as for semi-micro SEC applications. Moreover, the instrument, in combination with TSK-GEL SuperMultiporeHZ Series columns and PStQuick calibration standards, demonstrates a high level of robustness in terms of system-to-system reproducibility of molecular mass measurements.

- Combined with the TSK-GEL SuperMultiporeHZ semi-micro columns and the PStQuick calibration kits, the EcoSEC GPC system reduces solvent costs, increases throughput and minimizes preparation errors. The EcoSEC GPC system is an ideal instrument designed to meet the challenges of today's laboratory environment and demanding workload for GPC analysis.