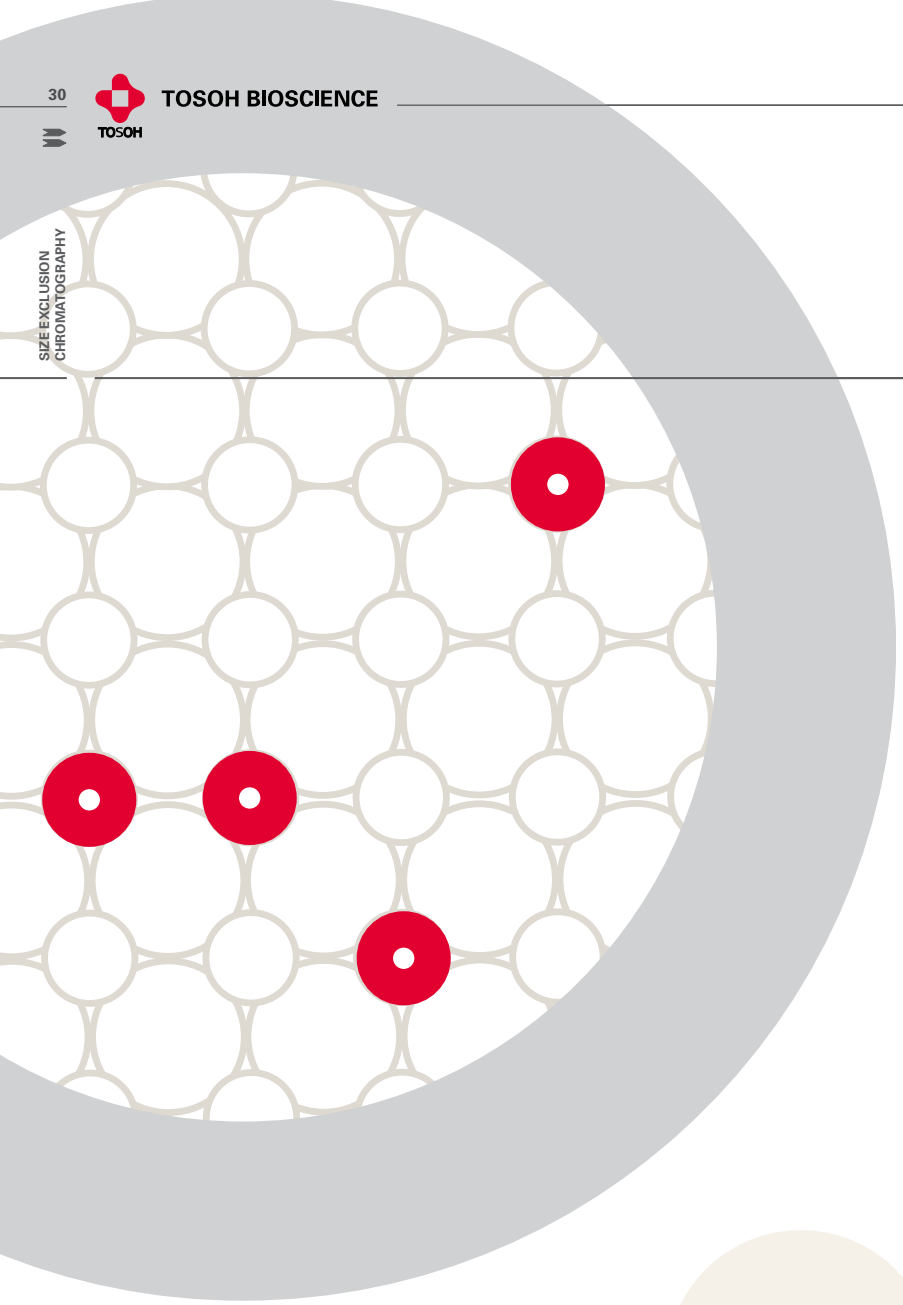
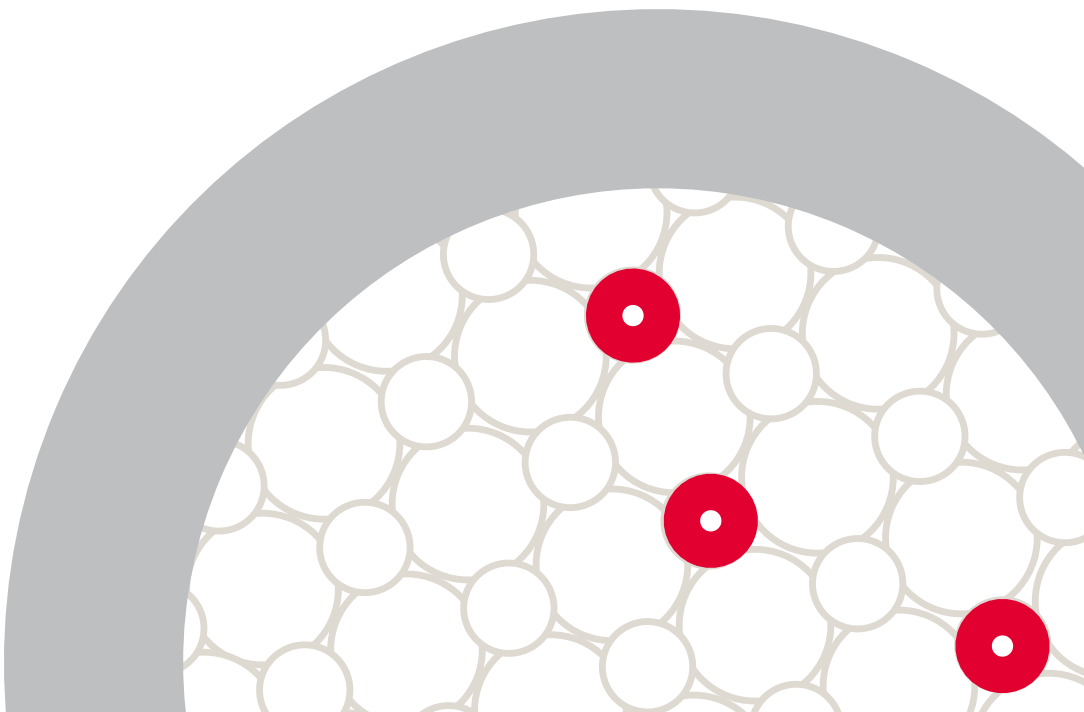




SEC
SIZE EXCLUSION CHROMATOGRAPHY

SEC PRODUCTS

TSKgel SW-type

TSKgel SW
TSKgel SWXL
TSKgel SuperSW

TSKgel PW-type

TSKgel PW
TSKgel PWXL
TSKgel PWXL-CP
TSkgel SuperMultipore PW
TSkgel SuperOligo PW

TSKgel Alpha-type

TSKgel Alpha
TSKgel SuperAW
TSKgel Vmpak

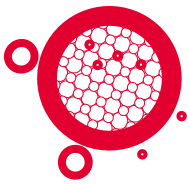
TSKgel H-type

TSKgel HXL
TSKgel HHR
TSKgel SuperH
TSKgel SuperHZ
TSKgel MultiporeHZ

TOSOH FACT

Tosoh has a long history in size exclusion chromatography (SEC). In 1978 Tosoh first introduced porous silica-based SW columns for the isolation of proteins using LC. These first gels had particle sizes from 10 to 13 µm and were quickly adopted and referred to as the standard for analytical SEC on FPLC and HPLC systems.

As new packing materials were discovered and new bonding chemistries developed, the SEC product line has grown into four major classes of SEC columns. The following pages will help you choose the best column for your application.



INTRODUCTION TO TSKgel SIZE EXCLUSION COLUMNS

GEL FILTRATION CHROMATOGRAPHY (GFC)
GFC is popular among biochemists for the isolation of proteins, for the removal of aggregates, to desalt a protein sample, to separate nucleic acid fractions, or to characterize water-soluble polymers used in food products, paints, pharmaceutical preparations, etc. Available TSKgel products are classified by application area and particle composition. Each of the types below is described in detail in this chapter.

Application Area: Proteins and other biopolymers

Base material: silica

- SW
- SWXL
- SuperSW

These columns are ideal for proteins and nucleic acids using an aqueous buffer as mobile phase.

Application Area: Water-soluble polymers

Base material: polymethacrylate

- PW
- SuperMultiporePW
- SuperOligoPW
- PWXL
- PWXL-CP

These columns are ideal for industrial polymers, oligosaccharides, nucleic acids and small viruses using aqueous buffer or salt solutions as mobile phase. The new TSKgel SuperMultiporePW semi-micro SEC columns provide near linear calibration curves and are ideally suited to analyze the MW distribution of water soluble polymers with a wide range of molecular weights. The SuperOligoPW semi-micro column featuring a small particle size has been designed for fast analysis of oligosaccharides and other oligomers. The PW_{XL}-CP columns are developed to facilitate SEC separation of cationic polymer under low salt conditions.

Application Area: Water- and organic-soluble polymers

Base material: highly crosslinked polymethacrylate

- Alpha
- SuperAW

These columns are ideal for industrial polymers soluble in water, buffers and many organic solvents.

GEL PERMEATION CHROMATOGRAPHY (GPC)
GPC plays an important role in the characterization of organic-soluble polymers in the chemical and petrochemical industries. TSKgel GPC columns contain particles prepared from polystyrene crosslinked with divinylbenzene.

The proprietary multi-pore particle technology applied in some linear GPC columns ensures a wide pore size distribution in each particle leading to calibration curves with excellent linearity. Available GPC columns are grouped according to their relative lack of adsorptive properties and the speed of analysis.

Each of the types below is described in detail in this chapter.

Application Area: Organic-soluble polymers

Base material: polystyrene

Ultra-low adsorption columns with limited solvent range

- SuperHZ (high throughput)
- SuperMultiporeHZ
- HXL (conventional)

Low adsorption columns with expanded solvent range

- SuperH (high throughput)
- HHR (conventional)

FEATURES

- Rigid hydrophilic and hydrophobic packings
- Four series of SEC columns with diferent ranges of solvent compatibility
- Easy scale up

BENEFITS

- Minimal swelling and excellent physical strength
- Low adsorption resulting in high mass recovery
- Suitable for both types of size exclusion, aqueous (GFC) and non-aqueous (GPC)
- Analytical and preparative pre-packed SEC column

SEC

SUMMARY OF TSKgel SIZE EXCLUSION COLUMN LINES

Column line	TSKgel SW / SWXL / SuperSW	TSKgel PW / PWXL	TSKgel Alpha / TSKgel SuperAW	TSKgel H
Particle composition	Silica	Polymethacrylate	highly crosslinked Poly-methacrylate	PS-DVB
No. of available pore sizes	3/2	7	5	6
pH stability	2.5 - 7.5	2.0 - 12.0	2.0 - 12.0	1.0 - 14.0
Solvent compatibility	100% polar	50% polar	100% polar and nonpolar	100% nonpolar, limited polar
Max. temperature	30°C	80°C*	80°C	60-80°C (HXL and SuperHZ) 140°C (HHR and SuperH)
Max flow rate (mL/min)	6.0 (SW, SWXL) / 0.4 (SuperSW)	1.2 (PW) / 1.0 (PWXL)	1.0 (Alpha) / 0.6 (SuperAW)	
Pressure (MPa)**	1.0-12.0	1.0 - 4.0	2.0 - 4.0	15-60
Application focus	proteins	water-soluble polymers	intermediate polar polymers	organic-soluble polymers

* *Except for the TSKgel G-DNA-PW, which can be operated up to 50°C and the 55 mm ID TSKgel PW-type columns, which can be operated up to 60°C. When operating below 10°C, reduce the flow rate to ensure that the maximum pressure is not exceeded.*

** *Depends on column dimensions and particle size.*

Note: The operating conditions and specifications for each column are listed on the Operating Conditions and Specifications sheet (OCS) shipped with the column and in the Ordering Information section at the end of each section.

COLUMN SELECTION GUIDE FOR TSKgel GEL FILTRATION COLUMNS

SAMPLE			COLUMN SELECTION		SELECTION CRITERIA
			FIRST CHOICE	ALTERNATIVE	
Carbohydrates	polysaccharides		TSKgel GMPW$_{XL}$ TSKgel SuperMultiporePW	TSKgel G5000PW$_{XL}$ & TSKgel G3000PW$_{XL}$	large pore size, small particles, linear calibration curve, high resolving power
	oligosaccharides		TSKgel G-Oligo-PW TSKgel SuperOligoPW	TSKgel G2500PW$_{XL}$	small particles, high resolving power
Nucleic acids	DNA fragments	large	TSKgel G-DNA-PW or TSKgel G5000PW$_{XL}$		large pore size, small particles, high resolving power
		medium and small	TSKgel G4000SW$_{XL}$, TSKgel BioAssist G4SW$_{XL}$ TSKgel SuperSW3000 or TSKgel G3000SW$_{XL}$ TSKgel BioAssist G3SW$_{XL}$		suitable pore sizes
	RNA		TSKgel G4000SW$_{XL}$ TSKgel BioAssist G4SW$_{XL}$ TSKgel SuperSW3000 or TSKgel G3000SW$_{XL}$ TSKgel BioAssist G3SW$_{XL}$		suitable pore sizes
	oligonucleotides		TSKgel G2500PW$_{XL}$		small pore size, ionic interaction
Proteins	normal size small-medium proteins		TSKgel SuperSW3000 TSKgel G3000SW$_{XL}$ TSKgel BioAssist G3SW$_{XL}$ TSKgel G4000SW$_{XL}$ TSKgel BioAssist G4SW$_{XL}$ TSKgel SuperSW2000 or TSKgel G2000SW$_{XL}$ TSKgel BioAssist G2SW$_{XL}$	TSKgel G3000PW$_{XL}$ / G4000PW$_{XL}$	small particles small to medium range pore sizes
	large proteins	low density lipoprotein	TSKgel G6000PW$_{XL}$ or TSKgel G5000PW$_{XL}$		large pore sizes
		gelatin	TSKgel GMPW$_{XL}$ TSKgel SuperMultiporePW-M TSKgel G3000SW$_{XL}$	TSKgel G5000PW$_{XL}$ & G3000PW$_{XL}$	large pore size, linear calibration curve
Peptides	large		TSKgel SuperSW3000 TSKgel G3000SW$_{XL}$ TSKgel BioAssist G3SW$_{XL}$ or TSKgel G2000SW$_{XL}$ TSKgel BioAssist G2SW$_{XL}$	TSKgel SuperSW2000 / TSKgel G3000PW$_{XL}$	small to medium range pore size, versatile
	small		TSKgel G2500PW$_{XL}$	TSKgel SuperSW2000 / TSKgel G2000SW$_{XL}$	linear calibration curve, high resolving power
Viruses			TSKgel G6000PW$_{XL}$ or TSKgel G5000PW$_{XL}$ TSKgel SuperMultiporePW-H		large pore size, high resolving power
Synthetic polymers			TSKgel GMPW$_{XL}$ or TSKgel Alpha-M TSKgel SuperMultiporePW	TSKgel G5000PW$_{XL}$ & G3000PW$_{XL}$ / TSKgel Alpha-5000 & Alpha-3000	large pore size, low adsorption, linear calibration curve
	cationic		TSKgel G3000PW$_{XL}$-CP TSKgel G5000PW$_{XL}$-CP TSKgel G6000PW$_{XL}$-CP		medium to large pore size, low adsorption, linear calibration curve
Synthetic oligomers	nonionic		TSKgel G-Oligo-PW TSKgel G2500PW$_{XL}$ or TSKgel Alpha-2500 TSKgel SuperOligoPW and TSKgel SuperMultiporePW-N	TSKgel G2500PW / TSKgel SuperAW2500	small pore size, high resolving power
	anionic		TSKgel G2500PW$_{XL}$ or TSKgel Alpha-2500	TSKgel G2500PW / TSKgel SuperAW2500	small pore size, ionic interaction

SEC

TSKgel SW, SWXL AND SUPERSW GEL FILTRATION COLUMNS

HIGHLIGHTS

- TSKgel SW-type columns are all based on spherical silica particles with very high internal pore volumes.
- Silica particles in SW-type columns are chemically bonded with polar diol groups.
- SW-type columns feature low residual adsorption, which is essential for gel filtration analysis.
- Three pore sizes ranges (125 Å, 250 Å and 450 Å) available.
- Stainless steel, glass and PEEK column hardware available.

TSKgel SW series columns (SW, SWXL and Super SW) contain a large pore volume per unit column volume, which results in either higher MW selectivity or better resolution when analyzing proteins. They are based on highly porous silica particles, the surface of which has been shielded from interacting with proteins by derivatization with ligands containing diol functional groups. TSKgel SW series columns stand out from other silica- or polymer-based high performance size exclusion columns by virtue of their large pore volumes and low residual adsorption.

SW and SWXL columns are available in three pore size ranges with nominal pore sizes of 125 Å, 250 Å and 450 Å. SuperSW and QC-PAK column lines are available in 125 Å and 250 Å. SW columns are packed with 10 micron (G2000SW and G3000SW) or 13 micron (G4000SW) particles. SWXL columns contain 5 micron (G2000SWXL and G3000SWXL) or 8 micron (G4000SWXL) particles. SuperSW columns contain 4 micron particles.

RECOMMENDATIONS FOR TSKgel SW SERIES SELECTION SAMPLES OF UNKNOWN MOLECULAR WEIGHT

TSKgel G3000SWXL is the ideal scouting column. If the protein of interest elutes near the exclusion volume, then G4000SWXL is the logical next step. Conversely, if the protein of interest elutes near the end of the chromatogram, try the G2000SWXL.

Proteins (general)

Choose one of the TSKgel SWXL columns using the calibration curves on page 34 to select the appropriate pore size based on knowledge or estimate of protein size.

Monoclonal antibodies

TSKgel G3000SWXL is commonly used for quality control. TSKgel SuperSW3000 is utilized when sample is limited or at very low concentration.

Peptides

TSKgel G2000SWXL is the first selection for the analysis of peptides. TSKgel SuperSW2000 is utilized when sample is limited or at very low concentration.

Other

The use of TSKgel SuperSW columns requires optimization of the HPLC system with respect to extra-column band broadening. Capillary tubing ID, injection volume, detector cell volume, and detector time constant all need to be reduced to fully benefit from the high column efficiency and small peak volumes of the SuperSW columns. Use SW columns when not sample limited or when larger amounts of sample need to be isolated.

TABLE 1

Properties and Separation Ranges for TSKgel SW-Type Packings

			Molecular weight of sample (Da)		
TSKgel packing	**Particle size (µm)**	**Pore size (Å)**	**Globular proteins**	**Dextrans**	**Polyethylene glycols and oxides**
SuperSW2000	4	125	$5 \times 10^3 – 1.5 \times 10^5$	$1 \times 10^3 – 3 \times 10^4$	$5 \times 10^2 – 15 \times 10^3$
G2000SWXL/BioAssist G2SWXL	5	125	$5 \times 10^3 – 1.5 \times 10^5$	$1 \times 10^3 – 3 \times 10^4$	$5 \times 10^2 – 15 \times 10^3$
QC-PAK TSK 200	5	125	$5 \times 10^3 – 1.5 \times 10^5$	$1 \times 10^3 – 3 \times 10^4$	$5 \times 10^2 – 15 \times 10^3$
G2000SW	10, 13, 20	125	$5 \times 10^3 – 1.5 \times 10^5$	$1 \times 10^3 – 3 \times 10^4$	$5 \times 10^2 – 15 \times 10^3$
SuperSW3000	4	250	$1 \times 10^4 – 5 \times 10^5$	$2 \times 10^3 – 7 \times 10^4$	$1 \times 10^3 – 3.5 \times 10^4$
G3000SWXL/BioAssist G3SWXL	5	250	$1 \times 10^4 – 5 \times 10^5$	$2 \times 10^3 – 7 \times 10^4$	$1 \times 10^3 – 3.5 \times 10^4$
QC-PAK TSK 300	5	250	$1 \times 10^4 – 5 \times 10^5$	$2 \times 10^3 – 7 \times 10^4$	$1 \times 10^3 – 3.5 \times 10^4$
G3000SW	10, 13, 20	250	$1 \times 10^4 – 5 \times 10^5$	$2 \times 10^3 – 7 \times 10^4$	$1 \times 10^3 – 3.5 \times 10^4$
G4000SWXL/BioAssist G4SWXL	8	450	$2 \times 10^4 – 7 \times 10^6$	$4 \times 10^3 – 5 \times 10^5$	$2 \times 10^3 – 2.5 \times 10^5$
G4000SW	13, 17	450	$2 \times 10^4 – 7 \times 10^6$	$4 \times 10^3 – 5 \times 10^5$	$2 \times 10^3 – 2.5 \times 10^5$

Data generated using the following conditions:

Columns: Two 4 µm, 4.6 mm ID x 30cm L TSKgel SuperSW columns in series; two 5 µm, 7.8 mm ID x 30 cm L TSKgel SWXL columns in series; two 10 µm, 7.5 mm ID x 60 cm L TSKgel SW columns in series

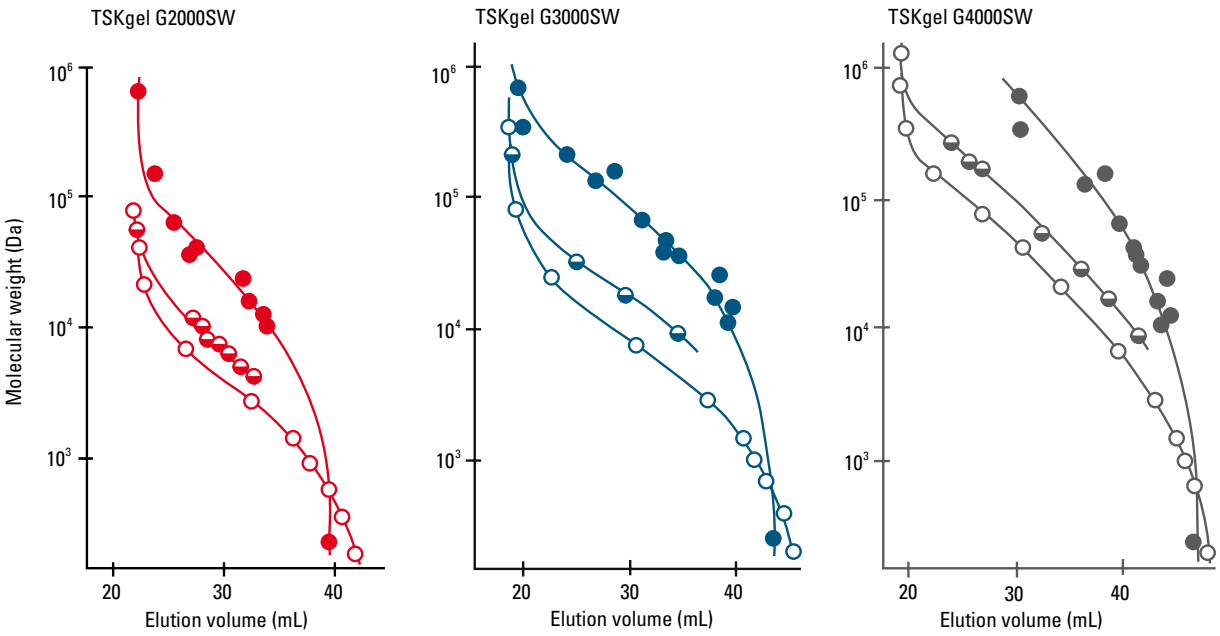
Elution: Globular proteins: 0.3 mol/L NaCl in 0.1 mol/L (0.05 mol/L for SWXL columns) phosphate buffer, pH 7.0

Dextrans and polyethylene glycols and oxides (PEOs): distilled water

CALIBRATION CURVES FOR TSKgel SW-TYPE GEL FILTRATION COLUMNS

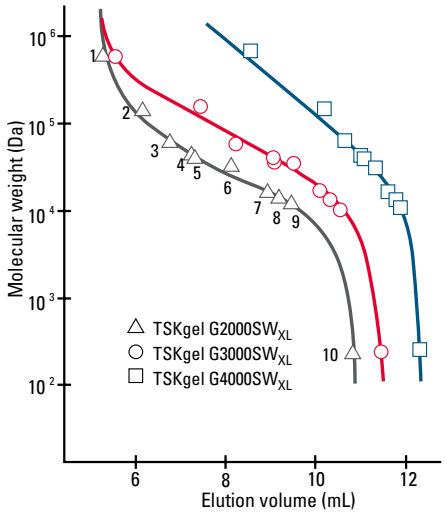
The best results are obtained when selecting a column with the sample's molecular weight in the linear portion of the calibration curve.

Polyethylene oxide, dextran and protein calibration curves for TSKgel SW columns



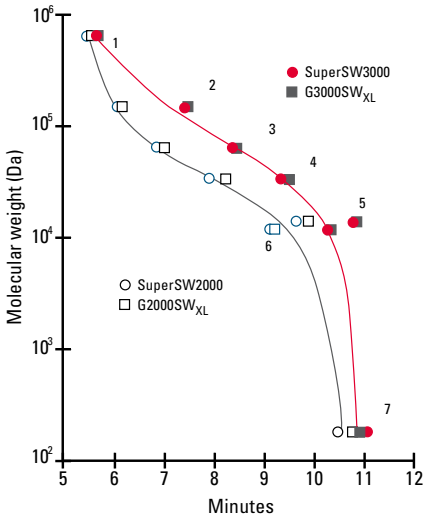
Column: TSK-GEL SW, two 7.5 mm ID x 60 cm L columns in series
Sample: ● proteins, ○ polyethylene oxides, ◒ dextrans
Elution: dextrans and polyethylene oxides: distilled water; proteins: 0.3 mol/L NaCl in 0.1 mol/L phosphate buffer, pH 7.0
Flow Rate: 1.0 mL/min
Detection: UV @ 220 nm and RI

Protein calibration curves for TSKgel SW$_{XL}$ columns



Column: TSK-GEL SW$_{XL}$ columns, 5 or 8 μm ,7.8 mm ID x 30 cm L
Sample: 1. thyroglobulin (660,000 Da); 2. IgG (160,000 Da);
3. BSA (67,000 Da); 4. ovalbumin (43,000 Da);
5. peroxidase (40,200 Da); 6. β-lactoglobulin (18,400 Da);
7. myoglobin (16,900 D a); 8. ribonuclease A (12,600 Da);
9. cytochrome C (12,400 Da); 10. glycine tetramer (246 Da)
Elution: 0.3 mol/L NaCl in 0.1 mol/L sodium phosphate buffer, pH 7.0
Detection: UV @ 220 nm

Calibration curves for TSKgel Super SW and SW$_{XL}$



Sample: proteins: 1. thyroglobulin (660,000 Da);
2. γ-globulin (150,000 Da); 3. BSA (67,000 Da);
4. β-lactoglobulin (18,400 Da); 5. lysozyme (14,500 Da);
6. cytochrome C (12,400 Da); 7. triglycine (189 Da)
Elution: 0.15 mol/L phosphate buffer (pH 6.8)
Flow Rate: 0.35 mL/min for SuperSW; 1.0 mL/min for SW$_{XL}$
Temperature: 25°C
Detection: UV @ 280 nm (220 nm for triglycine)

SEC

APPLICATIONS OF TSKgel SW-TYPE GEL FILTRATION COLUMNS

COMPARING TSKgel SW, SWXL AND SUPERSW GEL FILTRATION COLUMNS

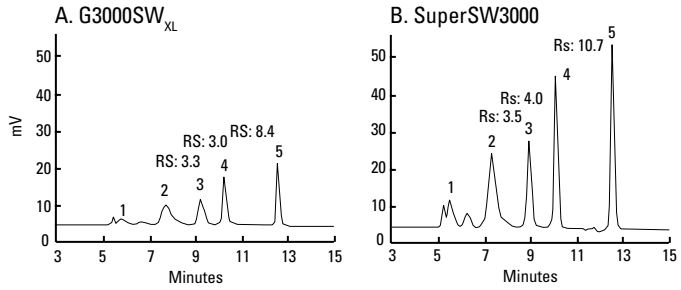
FIGURE 1 & FIGURE 2 show the increased resolution and sensitivity of the TSKgel SuperSW columns compared to TSKgel SWXL columns. This is due to the smaller particle size (4 vs. 5 µm) coupled with a narrow column (4.6 mm ID).

ANALYSIS OF PROTEIN AGGREGATION

TSKgel G3000SWXL columns are the industry standard for agregation analysis in quality control of monoclonal antibodies (mAbs). FIGURE 3 shows the analysis of mAb Aggregates with UV, refractive index (RI) and multi angle light scattering (MALS) detection. When the protein analysis needs to be performed in a metal free environment, the BioAssistSW series offers TSKgel SW packings in PEEK housings, featuring the same performance as stainless steel columns.

FIGURE 1

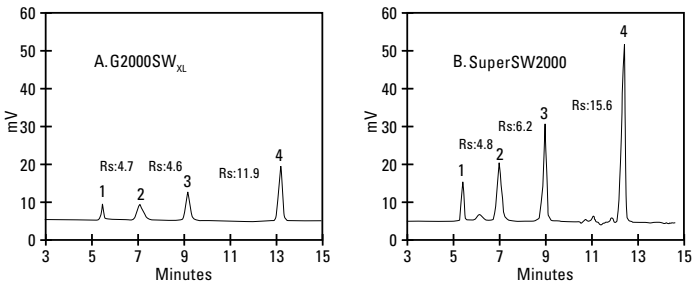
Comparison of TSKgel Super SW3000 and TSKgel G3000SWXL for the separation of proteins



Column: A. TSKgel G3000SWXL, 7.8 mm ID x 30 cm L;
B. TSKgel SuperSW3000, 4.6 mm ID x 30 cm L;
Sample: 5 µL of a mixture of 1. thyroglobulin, 0.5 mg/mL (660,000 Da); 2. γ-globulin, 1.0 mg/mL (150,000 Da); 3. ovalbumin, 1.0 mg/mL (43,000 Da); 4. ribonuclease A, 1.5 mg/mL (12,600 Da); 5. ρ-aminobenzoic acid, 0.01 mg/mL (137 Da);
Elution: 0.1 mol/L $NaSO_4$ in 0.1 mol/L in phosphate buffer with 0.05 % NaN_3, pH 6.7; Flow Rate: 1.0 mL/min for G3000SWXL; 0.35 mL/min for SuperSW3000;
Temp: 25°C; Detection: UV @ 220nm

FIGURE 2

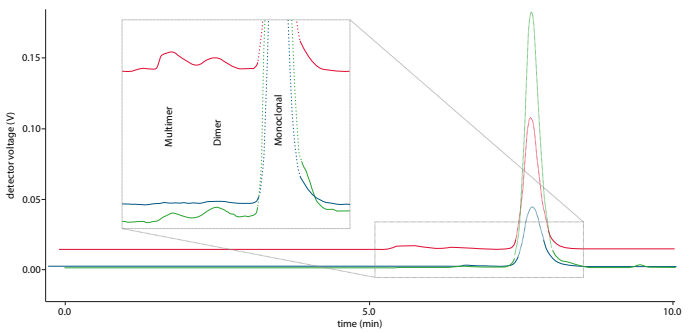
Comparison of TSKgel Super SW2000 and TSKgel G3000SW FOR the separation of Proteins



Column: A. TSKgel G2000SWXL, 7.8 mm ID x 30 cm L;
B. TSKgel SuperSW2000, 4.6 mm ID x 30cm L;
Sample: 1. thyroglobulin (0.2 mg/mL); 2. albumin (1.0 mg/mL); 3. ribonuclease A (1.0 mg/mL); 4. p-aminobenzoic acid (0.01 mg/mL); Inj. Volume: 5 µL;
Elution: 0.1 mol/L phosphate buffer + 0.1 mol/L Na_2SO_4 + 0.05 % NaN_3 (pH 6.7);
Flow Rate: 0.35 mL/min for SuperSW2000; 1.0 mL/min for G2000SWXL;
Temp: 25°C; Detection: UV @ 280nm

FIGURE 3

SEC-Mals-UV-RI analysis of MAB aggregates



Column: TSKgel G3000SWXL column, 5 µm, 7.8 mm ID x 30 cm L
Sample: monoclonal antibody, Inj.volume: 20 µL;
Mobile phase: phosphate buffered saline (PBS); Flow rate: 1 mL/min;
Detection: MALS (red), refractive index (blue) & UV @ 280 nm (green);
HPLC System: LC-20A prominence, Shimadzu;
MALS detector: miniDAWN™ TREOS, Wyatt Techn. Corp.

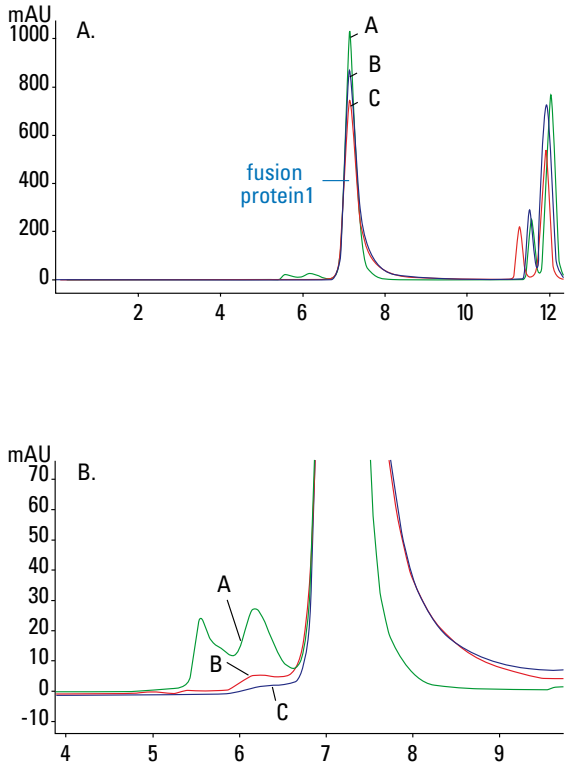
APPLICATIONS OF TSKgel SW-TYPE GEL FILTRATION COLUMNS

ANALYSIS OF ANTIBODY-FUSION PROTEINS

During method development, many variables are examined to ensure method robustness. Factors such as elution profile, peak shape, and recovery are required to be consistent. During a method re-qualification several variables were investigated to eliminate non-specific binding and increase the robustness of an established QC method using a TSKgel SuperSW3000 column. As shown in FIGURE 4, excessive peak tailing of "fusion protein 1" is evident with the use of 0.2 mol/L NaCl (chromatogram c). Additionally, the expected protein dimer and trimer aggregates are not visible. By switching from 0.2 mol/L sodium chloride to 0.2 mol/L of the more chaotropic sodium perchlorate salt, together with a two-fold reduction in the buffer concentration, less peak tailing and distinct peaks for the dimer and trimer species of mAb 1 resulted (chromatogram B). Doubling the perchlorate concentration to 0.4 mol/L provided further improvement in the peak shape of fusion protein 1 and associated aggregate species (chromatogram A). FIGURE 4B is an enlargement of the baseline region, showing an improved peak shape of the dimer and trimer aggregates with the use of 0.4 mol/L $NaClO_4$.

FIGURE 4

Overlays of Antibody fusion protein analysis



Column: TSKgel SuperSW3000, 4 µm, 4.6 mm ID x 30 cm L;
Mobile phase: c: 0.4 mol/L $NaClO_4$, 0.05 mol/L NaH_2PO_4, b: 0.2 mol/L $NaClO_4$, 0.05 mol/L NaH_2PO_4, a: 0.2 mol/L NaCl, 0.1 mol/L NaH_2PO_4; Flow rate: 0.35 mL/min; Detection: UV@214nm; Injection vol.: 5 µL; Samples: antibody fusion protein

MEMBRANE PROTEINS

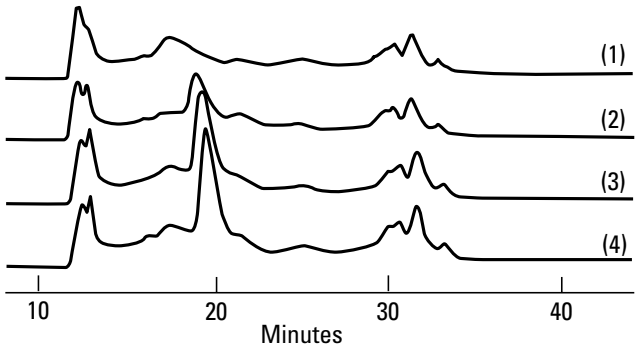
The effect of different concentrations of surfactant on the separation of membrane proteins is seen in FIGURE 5. As the concentration of octaethyleneglycol dodecylether increases to 0.05%, the main peak becomes sharper and recovery increases.

ENZYMES

Mobile phase conditions in GFC are optimized to ensure little or no interaction of the sample with the packing material. This gentle technique allows for high recovery of enzymatic activity. A crude sample of glutathione S-transferase was separated in only 15 minutes on a TSKgel G3000SWXL column and activity recovery was 98% and 89%, respectively. The elution profile of the separation in FIGURE 6 shows that all of the activity eluted in a narrow band of about 1.5 mL.

FIGURE 5

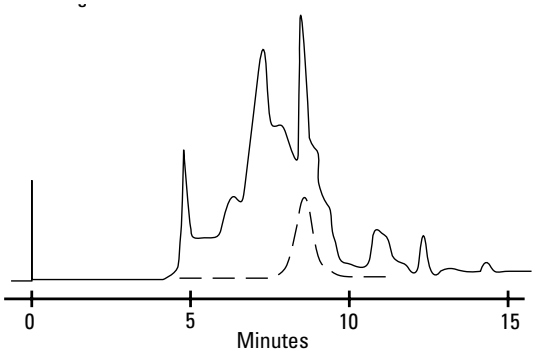
Separation of membrane protein by SEC with different surfactant concentration in the eluent



Column: TSKgel G3000SW, 7.5mm ID x 60 cm L; Sample: Membrane protein from a crude extract from rat liver microsome; Elution: (0.2 mol/L sodium chloride + 20 % glycerol + octaethyleneglycol dodecylether) in 50 mmol/L phosphate buffer, pH 7.0.
Note: concentration of surfactant: (1) 0.005 %, (2) 0.01 %, (3) 0.025 %, (4) 0.05 %;
Flow Rate: 1.0 mL/min; Detection: UV @ 280 nm

FIGURE 6

Separation of crude protein sample on TSKgel G3000SWXL



Column: TSKgel G3000SWXL 5 µm, (7.8 mm ID x 30 cm L); Sample: crude glutathione S-transferase from guinea pig liver extract, 0.7 mg in 0.1 mL;
Elution: 0.3 mol/L NaCl in 0.05 mol/L phosphate buffer, pH 7;
Flow Rate: 1.0mL/min; Detection: UV@220 nm (solid line) and enzyme assay tests (dashed line); Recovery: enzymatic activity recovered was 89 %

SEC

ORDERING INFORMATION

Part #	Description	ID (mm)	Length (cm)	Particle size (µm)	Number theoretical plates	Flow rate (mL/min) Range	Flow rate (mL/min) Max.	Maximum pressure drop (MPa)
TSKgel Glass columns								
16214	QC-PAK GFC 200GL	8.0	15	5	≥ 10,000	0.5 - 1.0	1.2	40
16216	QC-PAK GFC 300GL	8.0	15	5	≥ 10,000	0.5 - 1.0	1.2	40
08800	G3000SW, Glass	8.0	30	10	≥ 10,000	0.4 - 0.8	0.8	20
08801	G4000SW, Glass	8.0	30	13	≥ 8,000	0.4 - 0.8	0.8	20
TSKgel Stainless steel columns								
18674	SuperSW2000	4.6	30	4	≥ 30,000	0.1 -0.35	0.4	120
21845	SuperSW3000	1.0	30	4	≥ 18,000	0.016	0.02	120
21485	SuperSW3000	2.0	30	4	≥ 25,000	0.065	0.075	120
18675	SuperSW3000	4.6	30	4	≥ 30,000	0.1 - 0.35	0.4	120
08540	G2000SWXL	7.8	30	5	≥ 20,000	0.5 -1.0	1.2	70
08541	G3000SWXL	7.8	30	5	≥ 20,000	0.5 - 1.0	1.2	70
08542	G4000SWXL	7.8	30	8	≥ 16,000	0.5 - 1.0	1.2	35
16215	QC-PAK GFC 200	7.8	15	5	≥ 10,000	0.5 -1.0	1.2	40
16049	QC-PAK GFC 300	7.8	15	5	≥ 10,000	0.5 -1.0	1.2	40
05788	G2000SW	7.5	30	10	≥ 10,000	0.5 -1.0	1.2	20
05789	G3000SW	7.5	30	10	≥ 10,000	0.5 -1.0	1.2	25
05790	G4000SW	7.5	30	13	≥ 8,000	0.5 -1.0	1.2	15
05102	G2000SW	7.5	60	10	≥ 20,000	0.5 -1.0	1.2	40
05103	G3000SW	7.5	60	10	≥ 20,000	0.5 -1.0	1.2	50
05104	G4000SW	7.5	60	13	≥ 16,000	0.5 -1.0	1.2	30
06727	G2000SW	21.5	30	13	≥ 10,000	3.0 -6.0	8.0	10
06728	G3000SW	21.5	30	13	≥ 10,000	3.0 -6.0	8.0	15
06729	G4000SW	21.5	30	17	≥ 8,000	3.0 - 6.0	8.0	10
05146	G2000SW	21.5	60	13	≥ 20,000	3.0 -6.0	8.0	20
05147	G3000SW	21.5	60	13	≥ 20,000	3.0 -6.0	8.0	30
05148	G4000SW	21.5	60	17	≥ 16,000	3.0 -6.0	8.0	20
TSKgel PEEK Columns								
20027	BioAssist G2SWXL	7.8	30	5	≥ 20,000	0.5 - 1.0	1.2	70
20026	BioAssist G3SWXL	7.8	30	5	≥ 20,000	0.5 - 1.0	1.2	70
20025	BioAssist G4SWXL	7.8	30	8	≥ 16,000	0.5 - 1.0	1.2	35

ORDERING INFORMATION

Part #	Description	ID (mm)	Length (cm)	Particle size (µm)	
Guard column products					
08805	SW Guard column, Glass	8.0	4.0		For all 8 mm ID SW glass columns
18762	SuperSW Guard column	4.6	3.5		For 4.6 mm ID SuperSW columns (contains SuperSW3000 packing)
08543	SWXL Guard column	6.0	4.0		For all SWXL columns and P/Ns 16215 and 16049 (contains 3000SWXL packing)
18008	SWXL Guard column, PEEK	6.0	4.0		For all BioAssist SWXL, PEEK columns
05371	SW Guard column	7.5	7.5		For all 7.5 mm ID SW columns (contains 3000SW packing)
05758	SW Guard column	21.5	7.5		For all 21.5 mm ID SW columns
Bulk packing					
08544	SWXL Top-Off, 1g wet gel			5	For SWXL and QC-PAK columns
06819	SW Top-Off, 1g wet gel			10	For all 7.5 mm ID SW columns

SEC

TSKgel PW and TSKgel PWXL columns - Gel Filtration Chromatography of water soluble polymers

HIGHLIGHTS

- Hydrophilic, rigid, spherical, porous methacrylate beads pH range of 2 to 12, with up to 50% organic solvent
- Temperatures up to 80°C (50°C for TSKgel G-DNA-PW)
- Wide separation range up to 8 x 10^6 Da for linear polymers
- Linear SEC column line incorporating proprietary multi-pore technology
- Specialty columns for low salt separation of cationic polymers

Polymeric TSKgel PW and high resolution TSKgel PWXL columns are designed for SEC of water soluble organic polymers, polysaccharides, DNA, and RNA. They are based on a hydrophilic polymethacrylate matrix. For analytical purposes the TSKgel PWXL columns are preferred, whereas for preparative work the 60 cm TSKgel PW columns are recommended because of their higher loading capacity. For the analysis of proteins and peptides we recommend to use silica based SW type columns.

A number of specialty columns include columns for samples with a broad molecular weight range, oligosaccharides, DNA and RNA. A large pore G6000PW phase is available in PEEK column hardware (TSKgel BioAssist G6PW) for ultra-low sample adsorption during virus analysis. TSKgel PWXL-CP columns are especially suited for the separation of cationic polymers.

The latest addition to the TSKgel PW family are high resolution semi micro columns for oligomer analysis (TSKgel SuperOligoPW) and for analysis of MW distribution of by linear SEC (TSKgel SuperMultipore PW).

TABLE 2

Properties and Separation Ranges for TSKgel PW-Type Packings

TSKgel Column	Particle size (µm)	Pore size (Å)	MW range (PEG/PEO)	MW range Dextrans*
G1000PW		12	< 100	< 1 x 10^3
G2000PW	12	125	< 2 x 10^3	
G2500PW	12, 17	< 200	< 3 x 10^3	< 3 x 10^3
G3000PW	12, 17	200	< 5 x 10^4	
G4000PW	17	500	< 3 x 10^5	
G5000PW	17	1,000	< 1 x 10^6	
G6000PW/ BioAssist G6PW	17	> 1,000	< 8 x 10^6	
GMPW	17	< 100 - 1,000	5 x 10^2 - 8 x 10^6	
G2500PWXL	7	< 200	< 3 x 10^3	
G3000PWXL	7	200	< 5 x 10^4	< 6 x 10^4
G4000PWXL	10	< 500	< 3 x 10^5	1 x 10^3 - 7 x 10^5
G5000PWXL	10	1000	< 1 x 10^6	5 x 10^4 - 2.5 x 10^6
G6000PWXL	13	> 100	< 8 x 10^6	5 x 10^5 - 5 x 10^7
G-DNA-PW	10	> 1,000	< 8 x 10^6	< 5 x 10^7
GMPWXL	13	100 - 1,000	5 x 10^2 - 8 x 10^6	< 5 x 10^7
G-Oligo-PW	7	125	< 5 x 10^3	
SuperMultiporePW-N	4	n/a	3 x 10^2 - 5 x 10^4	
SuperMultiporePW-M	5	n/a	5 x 10^2 - 1 x 10^6	
SuperMultiporePW-H	8 (6-10)	n/a	1 x 10^3 - 1 x 10^7	
SuperOligoPW	3	n/a	1 x 10^2 - 3 x 10^3	
G3000PWXL-CP	7	200	< 9 x 10^4	
G5000PWXL-CP	10	1,000	< 1 x 10^6	
G6000PWXL-CP	13	> 1,000	< 2 x 10^7	

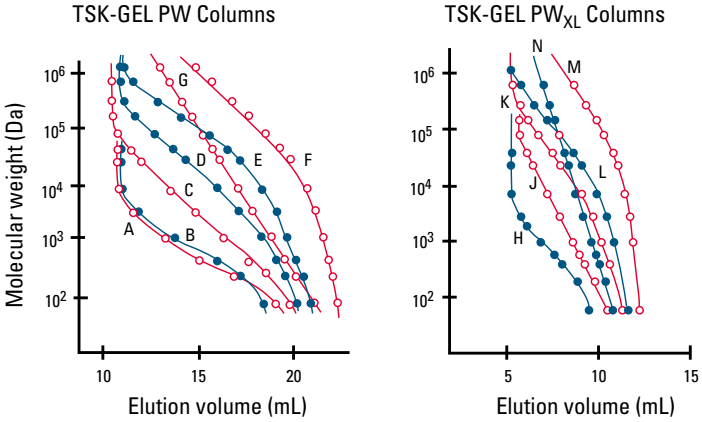
Column: TSKgel PW columns, 7.5 mm ID x 60 cm L; TSKgel PWXL, TSKgel PWXL-CP, G-Oligo-PW & G-DNA-PW, 7.8 mm ID x 30 cm L
Elution: Polyethylene glycols and oxides: distilled water; dextrans: 0.2 mol/L phosphate buffer, pH 6.8
Flow rate: 1.0 mL/min, except for TSKgel SuperMultiporePW and TSKgel SuperOligoPW columns: 0.6 mL/min
*Note: *Maximum separation range determined from estimated exclusion limits*

CALIBRATION CURVES FOR TSKgel PW / SUPERMULTIPORE PW GEL FILTRATION COLUMNS

The best results are obtained when selecting a column with the sample's molecular weight in the linear portion of the calibration curve.

FIGURE 7

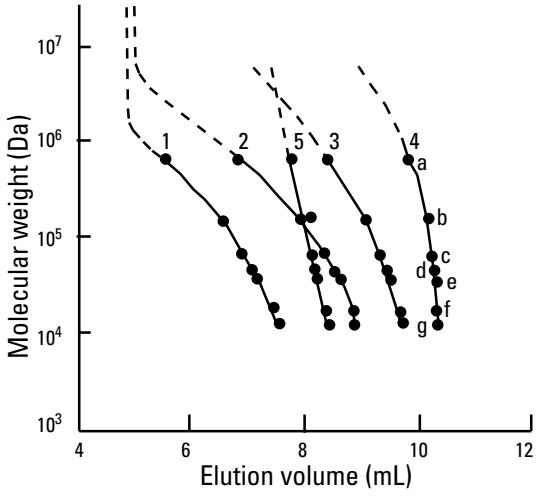
Polyethylene glycol and oxide calibration curves on TSKgel PW and TSKgel PWXL columns



Column: TSKgel PW columns: A. G2000PW, B. G2500PW, C. G3000PW, D. G4000PW, E. G5000PW, F. G6000PW, G. GMPW, all 7.5mm ID x 60 cm L TSKgel PWXL columns: H. G2500PWXL, J. G3000PWXL, K. G4000PWXL, L. G5000PWXL, M. G6000PWXL, N. GMPWXL, all 7.8 mm ID x 30 cm L; Elution: distilled water; Flow Rate: 1.0 m L/min; Detection: RI

FIGURE 8

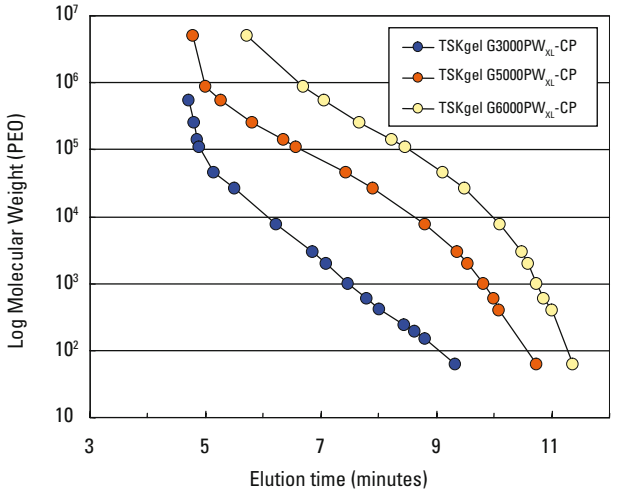
Protein calibration curves on TSKgel PWXL columns



Column: 1. TSKgel G3000PWXL, 2. G4000PWXL, 3. G5000PWXL, 4. G6000PWXL, 5. GMPWXL; Sample: a. thyroglobulin (660,000 Da), b. γ-globulin (150,000 Da), c. albumin (67,000 Da), d. ovalbumin (43,000 Da), e. β-lactoglobulin (36,000 Da), f. myoglobin (16,900 Da), g. cytochrome C (12,400 Da); Elution: 0.2 mol/L phosphate buffer (pH 6.8); Flow Rate: 1.0 mL/min; Detection: UV @ 280 nm

FIGURE 9

Polyethylene Glycol and Oxide Calibration Curves for TSKgel PWXL-CP Columns

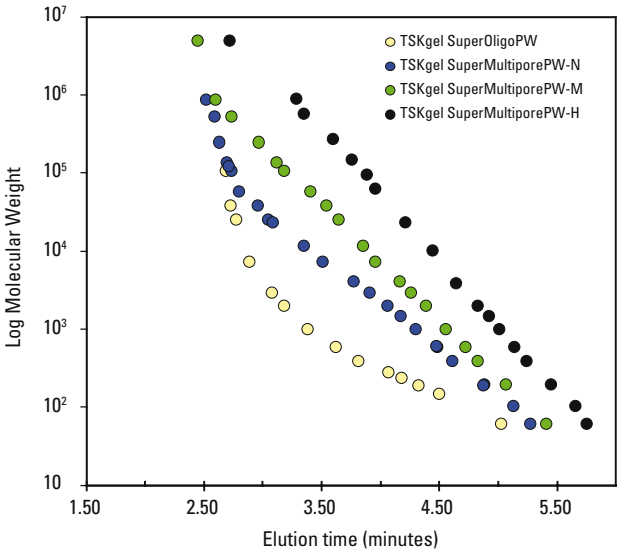


Columns: TSKgel G3000PWXL-CP, 7 µm, 7.8 mm ID x 30 cm L, TSKgel G5000PWXL-CP, 10 µm, 7.8 mm ID x 30 cm L, TSKgel G6000PWXL-CP, 13 µm, 7.8 mm ID x 30 cm L

Mobile phase: 0.1 mol/L $NaNO_3$; Flow Rate: 1 mL/min; Detection: RI; Temperature: 25°C; Samples: polyethylene oxides (PEO) standards, polyethylene glycols (PEG) standards

FIGURE 10

Polyethylene Glycol, Oxide and Ethylene Glycol Calibration Curves for TSKgel SuperMultiporePW and SuperOligoPW



Columns: TSKgel SuperOligoPW, SuperMultiporePW-N, SuperMultiporePW-M, SuperMultiporePW-H (each 6.0 mm ID x 15 cm L);

Mobile phase: H_2O; Flow rate: 0.60 mL/min; Detection: RI; Temperature: 25°C; Samples: polyethylene oxides (PEO) standards, polyethylene glycols (PEG) standards, ethylene glycol (EG) standards

SEC

COLUMNS FOR SPECIFIC APPLICATIONS

TSKgel PW$_{XL}$-CP

The new TSKgel PW$_{XL}$-CP columns are designed to facilitate the separation of cationic polymers by SEC at low salt conditions. They are based on the well known PW-type of polymeric resins for aqueous SEC. Cationic surface modification enables low salt elution of cationic polymers with high recoveries. The columns show high theoretical plate numbers, linear calibration curves and high durability. They are produced with three pore sizes for diffrent ranges (G3000-, G5000- and G6000PW$_{XL}$-CP). FIGURE 11 shows the analysis of various cationic polymers on a series of TSKgel PW$_{XL}$-CP columns.

TSKgel SUPEROLIGOPW & G-OLIGO-PW

The new TSKgel SuperOligoPW column was developed for the fast determination of molecular mass of aqueous oligomers, particularly oligosaccharides, and low molecular weight aqueous polymers. This is a semi-micro column (6.0 mm ID x 15 cm L) packed with spherical monodisperse polymethacrylate 3 µm particles. The combination of the decreased particle size and small dimensions of the TSKgel SuperOligoPW column enables high speed separation with high resolution - half of the separation time with the same resolution compared to conventional size exclusion columns. An added benefit of the semi-micro and small particle size is lower solvent consumption compared to conventional columns.

TSKgel G-Oligo-PW was designed for high resolution separations of nonionic and cationic oligomers and oligosaccharides such as hydrolyzed cyclodextrins. Because of the presence of residual cationic groups, this column is not recommended for separating anionic materials. The polyethylene glycol and polythylene oxide calibration curves for TSKgel G-Oligo-PW (not shown) are identical to the calibration curve for TSKgel G2500PW$_{XL}$ (shown on the previous page). FIGURE 12 shows the calibration curve for double stranded DNA for the TSKgel G-DNA-PW column.

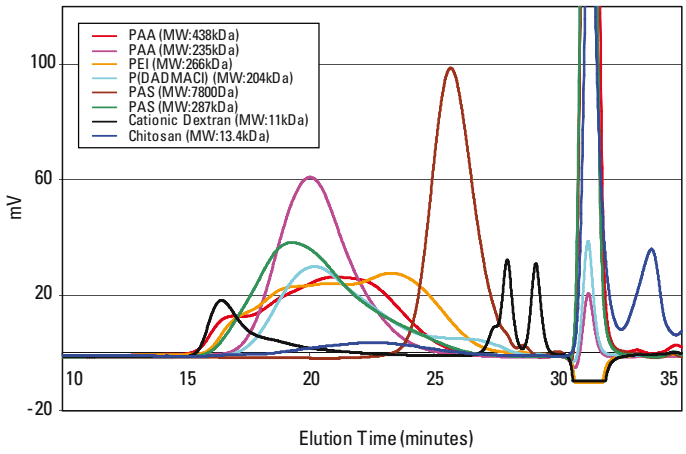
TSKgel G-DNA-PW

The TSKgel G-DNA-PW column is dedicated to the separation of large polynucleotides, such as DNA and RNA fragments of 500 to 5,000 base pairs. The exclusion limits for double-stranded DNA fragments are lower than those for rRNAs. The packing of the TSKgel G-DNA-PW column has very large pores (>1000 Å) and a small particle size (10 µm).

For the separation of large DNA fragments greater than 1,000 base pairs, a four-column system is typically required. Baseline resolution of DNA fragments up to 7,000 base pairs can be achieved, provided there is a two-fold difference in the chain length of the fragments.

FIGURE 11

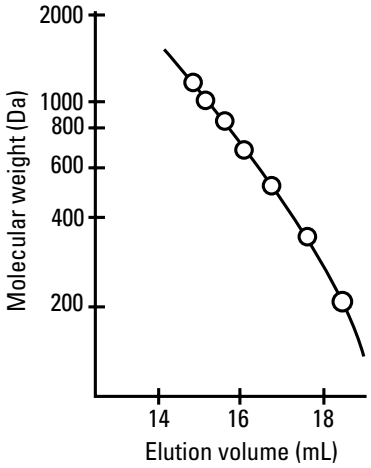
Double Stranded DNA Calibration Curve for TSKgel G-DNA-PW Column



Columns: TSKgel G3000PW$_{XL}$-CP, 7 µm (7.8 mm ID x 30 cm L), TSKgel G5000PW$_{XL}$-CP, 10 µm (7.8 mm ID x 30 cm L), TSKgel G6000PW$_{XL}$-CP, 13 µm (7.8 mm ID x 30 cm L); Eluent: 0.1 mol/L $NaNO_3$; Flow Rate: 1 mL/min; Detection: RI; Temperature: 25°C; Sample Load: 3 g/L, 100 µL

FIGURE 12

Oigosaccharides Calibration Curve for TSKgel G-Oligo-PW Column



Column: TSKgel G-Oligo-PW , two 6 µm, 7.8mm ID x 30cm L columns in series; Mobile phase: distilled H_2O; Flow Rate: 1.0 mL/min; Detection: UV@260 nm; Sample: hydrolyzed β-cyclodextrin

COLUMNS FOR SPECIFIC APPLICATIONS

TSKgel GMPW AND TSKgel GMPWXL

When the molecular weight range of the sample is broad or unknown, Tosoh Bioscience offers mixed-bed and multipore columns for analysis. The mixed bed column TSKgel GMPW and its high resolution counterpart, TSKgel GMPWXL, are packed with the G2500, G3000 and G6000 PW or corresponding PWXL resins. They offer a broad molecular weight separation range. As shown on page 42, the calibration curve for polyethylene glycols and oxides on these columns is fairly shallow and is linear over the range of 100-1,000,000 Da. The introduction of mixed-bed columns has facilitated the analysis of polydisperse samples. Previously, two-column systems such as TSKgel G3000PW and TSKgel G6000PW, were required to achieve good resolution with wide MW-range samples. The substitution of a TSKgel GMPW series column can save both time and money compared with multi-column systems.

TSKgel SuperMultiporePW

TSKgel SuperMultiporePW columns incorporate the multi-pore particle synthesis technology developed by Tosoh scientists in which monodisperse particles exhibit a broad range of pore sizes. See page 54 for additional information on multi-pore technology. Each particle, by design, has an extended linear calibration curve, thereby greatly diminishing the appearance of chromatograms with inflection points. This allows better reproducibility when determining molecular mass and molecular mass distribution of polymers.

Three semi-micro (6.0 mm ID x 15 cm L) columns are available within the TSKgel SuperMultiporePW series containing 4, 5 or 8 µm particles. This enables high speed separation for aqueous polymers and low solvent consumption compared to the conventional SEC columns. In addition, a wide separation range can be analyzed with the three different columns, from high molecular mass aqueous polymers to oligomers.

Multi-pore, semi-micro SEC columns provide high resolution and smooth peak shapes without shoulders of inflection points. This leads to better accuracy and reproducibility when determining the molecular mass distribution of water soluble polymers

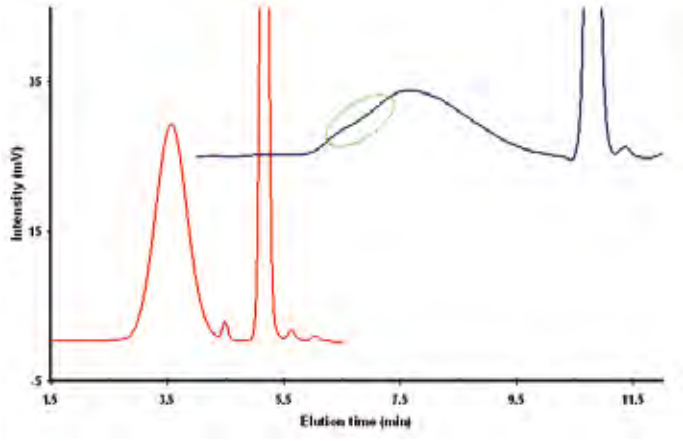
COMPARISON WITH CONVENTIONAL GPC COLUMNS

Figure 13 shows the SEC analysis of a real sample -Polyvinylpyrrolidone (PVP) K-30- on a series of conventional TSKgel G3000PWXL and G5000PWXL columns compared to the one obtained with a single TSKgel SuperMultiporePW-M linear SEC column (MW range 600,000 – 1,500,000). On a series of conventional SEC columns the Polyvinylpyrrolidone peak shows an inflection point, which does not appear on the SuperMultiporePW-M column. Analysis is much faster and more sensitive when applying the new multi-pore packing.

A mixture of polyethylene oxide (PEO) and polyethylene glycol (PEG) was analyzed on a semi-micro TSKgel SuperMultiporePW-M column and on conventional-sized TSKgel G3000PWXL and TSKgel G5000PWXL columns in series. As shown in Figure 14, the analysis using the TSKgel SuperMultiporePW-M column was completed in half the time and with higher resolution than the analysis performed using the TSKgel G3000PWXL and TSKgel G5000PWXL columns. This is due to the semi-micro dimensions (6.0 mm ID x 15 cm L) and the smaller particle size (5 µm) of the TSKgel SuperMultiporePW-M column compared to the 7.8 mm ID x 30 cm L size and 7 and 10 µm particle size of the TSKgel G3000PWXL and TSKgel G5000PWXL columns respectively.

FIGURE13

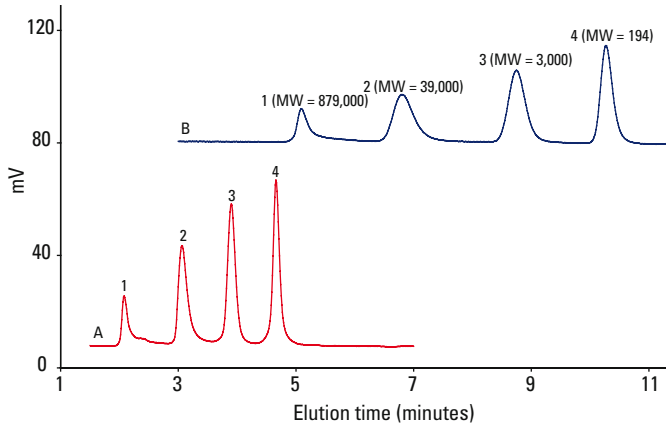
Analysis of Polyvenylpyrrolidone



Columns: TSKgel SuperMultiporePW-M, 6 mm ID x 15 cm L x 1 (red) TSKgel G3000PWXL & G5000PWXL, each 7.8 mm ID x 30 cm L in line (blue); Sample: Polyvinylpyrrolidone (K-30); Mobile phase: 0.1 mol/L $NaNO_3$; Flow rate: 0.6 mL/min; Detection: RI

FIGURE14

Comparison of analysis of a mixture of PEO and PEG



Column: TSKgel SuperMultiporePW-M, 6.0 mm ID x 15 cm L; TSKgel G5000PWXL + G3000PWXL, each 6.0 mm ID x 15 cm L; Mobile phase: H_2O; Flow rate: 0.6 mL/min; Detection: RI; Temperature: 25°C; Injection vol.: A: 20 µL, B: 100 µL; Samples: mixture of PEO and PEG

SEC

OPTIMIZING GEL FILTRATION WITH TSKgel PW AND TSKgel PWXL COLUMNS

SELECTING MOBILE PHASE BUFFERS

SEC separation is based on the difference of apparent molecular size with no additional interaction between the column matrix and the sample molecules. In practice, however, a small number of weakly charged groups on the surface of PW-type packings can cause changes in elution order from that of an ideal system. The eluent composition can vary greatly with TSKgel PW columns to be compatible with a wide range of neutral, polar, anionic, and cationic samples. The table below lists appropriate eluents for GFC of major polymer types.

For some nonionic, nonpolar polymers, such as polyethylene glycols, ideal size exclusion behavior can be obtained by using distilled water. More polar ionic polymers may exhibit abnormal peak shapes or minor peaks near the void volume when eluted with distilled water, due to ionic interactions between the sample and residual charged groups on the resin surface. To eliminate ionic interactions, a neutral salt such as sodium nitrate or sodium sulfate should be added. Generally, a salt concentration of 0.1 to 0.5 mol/L is sufficient to overcome undesirable ionic interactions.

HYDROPHOBIC SAMPLES

TSKgel PW-type resins are more hydrophobic than polysaccharide gels such as cross-linked dextran. Depending on the sample, this can lead to hydrophobic interaction as a secondary retention mechanism. The extent of hydrophobic interaction increases as the salt concentration of the eluent increases, but it can be reduced by the addition of an organic modifier such as acetonitrile. Water-soluble organic solvents are frequently used as modifiers to suppress hydrophobic interactions between the sample and the resin surface.

Modifiers are also used for optimizing the elution of both charged and neutral hydrophobic polymers. Typical examples for a variety of sample types are given in the table below. All TSKgel PW-type column packings are compatible with 20 % aqueous solutions of methanol, ethanol, propanol, acetonitrile, dimethyl formamide, dimethyl sulfoxide, formic acid, and acetic acid. In addition, these columns can be operated in 50 % aqueous acetone.

TABLE 3

Recommended eluents for GFC of water-soluble polymer on TSKgel PW-type columns

Type of polymer	Typical sample	Suitable eluent
Nonionic hydrophilic	polyethylene glycol soluble starch, methyl cellulose, pullulan dextran, hydroxyethyl cellulose, polyvinyl alcohol, polyacrylamide	distilled water 0.01N NaOH 20% DMSO Buffer or salt solution (e.g., 0.1–0.5 mol/L $NaNO_3$)
Nonionic hydrophobic	polyvinylpyrrolidone	Buffer or salt solution with organic solvent (e.g., 20% ACN in 0.1mol/L $NaNO_3$)
Anionic hydrophilic	sodium chondroitin sulfate, sodium alginate, carboxymethyl cellulose, sodium polyacrylate, sodium hyaluronate	Buffer or salt solution (e.g., 0.1 mol/L $NaNO_3$)
Anionic hydrophobic	sulfonated lignin sodium salt, sodium polystyrenesulfonate	Buffer or salt solution with organic solvent (e.g., 20% ACN in 0.1 mol/L $NaNO_3$)
Cationic hydrophilic	glycol chitosan, DEAE-dextran, poly(ethyleneimine), poly(trimethylaminoethyl methacrylate) iodide salt	0.5 mol/L acetic acid with 0.3 mol/L Na_2SO_4, or 0.8 mol/L $NaNO_3$ (0.1 mol/L $NaNO_3$ for PWXL-CP type)
Cationic hydrophobic	poly(4-vinylbenzyltrimethylammonium chloride), poly(N-methyl-2-vinylpyridinium) iodide salt	0.5 mol/L acetic acid with 0.3 mol/L Na_2SO_4
Amphoteric hydrophilic	peptides, proteins, poly-and oligosaccharides, DNA, RNA	Buffer or salt solution (e.g., 0.1 mol/L $NaNO_3$)
Amphoteric hydrophobic	blue dextran, collagen, gelatin, hydrophobic proteins hydrophobic peptides	Buffer or salt solution with organic solvent (e.g., 20% ACN in 0.1 mol/L $NaNO_3$ or 35 - 45% ACN in 0.1% TFA)

APPLICATIONS OF TSKgel PW-TYPE GEL FILTRATION COLUMNS

POLYSACCHARIDES

TSKgel PW columns are recommended for polysaccharide analysis due to their ability to separate a wide molecular weight distribution. Nonionic polysaccharides are the least complicated molecules to analyze by SEC because they seldom exhibit secondary interactions with the solid support. TSKgel G5000PW and TSKgel G3000PW in series are effective for the characterization of clinical dextran.

Cationic samples can be adsorbed on the resin by electrostatic interaction. If the polymer is strongly cationic, a fairly high salt concentration is required to prevent ionic interactions with conventional SEC packings. A mobile phase of 0.5 mol/L acetic acid with 0.3 mol/L Na_2SO_4 can also be used.

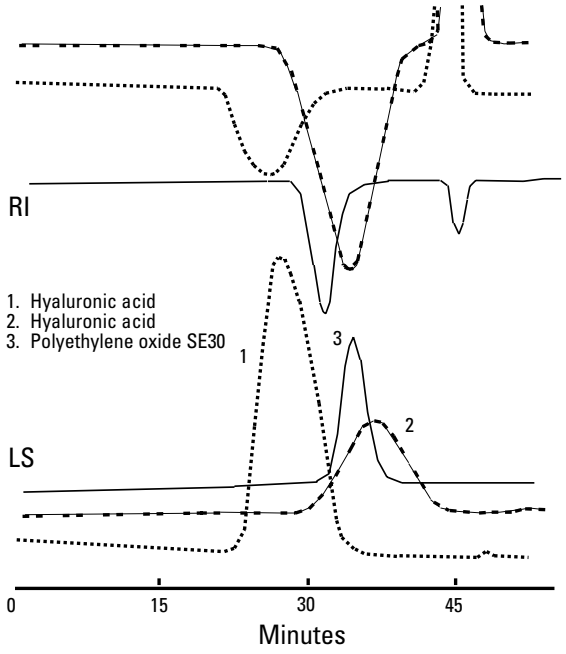
The new TSKgel PW$_{XL}$-CP series enables elution of water soluble, cationic polymers under low salt conditions (e.g. 0.1 mol/L $NaNO_3$). An effective separation of the anionic hydrophilic gluco-saminoglycan, hydraluronic acid, is shown in FIGURE 15 on a TSKgel G6000PW and TSKgel G4000PW column in series with a 0.2 mol/L sodium chloride mobile phase.

OLIGOSACCHARIDES

Figure 16 shows the rapid analysis of maltose oligomers using a TSKgel SuperOligoPW column compared to a TSKgel G-Oligo-PW column. The faster analysis time is due to the semi-micro dimensions (6.0 mm ID x 15 cm L) and the small particle size (3 µm) of the TSKgel SuperOligoPW column compared to the 7.8 mm ID x 30 cm L size and 7 µm particle size of the TSKgel G-Oligo-PW column.

FIGURE15

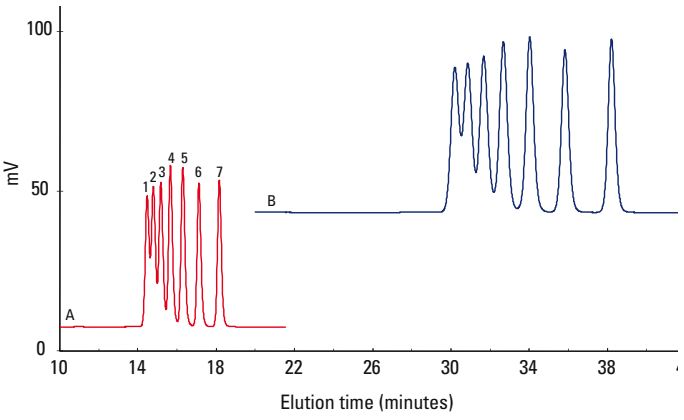
Analysis of Oligosaccharides



Column: TSKgel G6000PW + G4000PW, two 7.5 mm ID x 60 cm L columns in series; Mobile phase: 0.2 mol/L NaCl; Flow Rate: 0.9 mL/min
Temperature: 40°C; Samples: hyaluronic acid

FIGURE16

Analysis of Maltose Oligomers



Column: A: TSKgel SuperOligoPW, 3 µm, 6.0 mm ID x 15 cm L x 4
B: TSKgel G-Oligo-PW, 7 µm, 7.8 mm ID x 30 cm L x 4; Mobile phase: H_2O
Flow rate: A: 0.6 mL/min B: 1.0 mL/min; Detection: RI; Temperature: 40°C
Injection vol.: A: 10 µL B: 50 µL; Samples: 1.maltoheptose, 2. maltohexose, 3. maltopentose, 4. maltotetraose, 5. maltotriose, 6. maltose, 7. glucose

SEC

ORDERING INFORMATION

Part #	Description	ID (mm)	Length (cm)	Particle size (µm)	Number theoretical plates	Flow rate (mL/min) Range	Max.	Maximum pressure drop (MPa)
TSKgel Stainless Steel Columns								
22789	SuperMultiporePW-N	6.0	15	4	>16,000	0.3 - 0.6	0.6	4.5
22790	SuperMultiporePW-M	6.0	15	5	>12,000	0.3 - 0.6	0.6	2.7
22791	SuperMultiporePW-H	6.0	15	8 (6-10)	>7,000	0.3 - 0.6	0.6	0.9
22792	SuperOligoPW	6.0	15	3	>16,000	0.3 - 0.6	0.6	5.0
08031	G-Oligo-PW	7.8	30	7	≥ 16,000	0.5 - 0.8	1.0	4.0
08032	G-DNA-PW	7.8	30	10	≥ 10,000	0.2 - 0.5	0.6	2.0
08020	G2500PW$_{XL}$	7.8	30	7	≥ 16,000	0.5 - 0.8	1.0	4.0
08021	G3000PW$_{XL}$	7.8	30	7	≥ 16,000	0.5 - 0.8	1.0	4.0
08022	G4000PW$_{XL}$	7.8	30	10	≥ 10,000	0.3 - 0.6	1.0	2.0
08023	G5000PW$_{XL}$	7.8	30	10	≥ 10,000	0.3 - 0.6	1.0	2.0
08024	G6000PW$_{XL}$	7.8	30	13	≥ 7,000	0.3 - 0.6	1.0	2.0
08025	GMPW$_{XL}$	7.8	30	13	≥ 7,000	0.3 - 0.6	1.0	2.0
21873	G3000PW$_{XL}$-CP	7.8	30	7	≥ 16,000		1.0	5.5
21874	G5000PW$_{XL}$-CP	7.8	30	10	≥ 10,000		1.0	2.5
21875	G6000PW$_{XL}$-CP	7.8	30	13	≥ 7,000		1.0	2.0
05760	G1000PW	7.5	30	12	≥ 5,000	0.5 - 1.0	1.2	2.0
05761	G2000PW	7.5	30	12	≥ 5,000	0.5 - 1.0	1.2	2.0
08028	G2500PW	7.5	30	12	≥ 5,000	0.5 - 1.0	1.2	2.0
05762	G3000PW	7.5	30	12	≥ 5,000	0.5 - 1.0	1.2	2.0
05763	G4000PW	7.5	30	17	≥ 3,000	0.5 - 1.0	1.2	1.0
05764	G5000PW	7.5	30	17	≥ 3,000	0.5 - 1.0	1.2	1.0
05765	G6000PW	7.5	30	17	≥ 3,000	0.5 - 1.0	1.2	1.0
08026	GMPW	7.5	30	17	≥ 3,000	0.5 - 1.0	1.2	1.0
05105	G2000PW	7.5	60	12	≥ 10,000	0.5 - 1.0	1.2	4.0
08029	G2500PW	7.5	60	12	≥ 10,000	0.5 - 1.0	1.2	4.0
05106	G3000PW	7.5	60	12	≥ 10,000	0.5 - 1.0	1.2	4.0
05107	G4000PW	7.5	60	17	≥ 6,000	0.5 - 1.0	1.2	2.0
05108	G5000PW	7.5	60	17	≥ 6,000	0.5 - 1.0	1.2	2.0
05109	G6000PW	7.5	60	17	≥ 6,000	0.5 - 1.0	1.2	2.0
08027	GMPW	7.5	60	17	≥ 6,000	0.5 - 1.0	1.2	2.0
08030	G2500PW	21.5	60	17	≥ 10,000	1.6 - 6.0	8.0	2.0
PEEK								
20024	BioAssist G6PW	7.8	30	17	≥ 3,000	0.5 - 1.0	1.2	10
Guard columns								
22793	SuperMP (PW)-N Guard column	4.6	3.5	4				
22794	SuperMP (PW)-M Guard column	4.6	3.5	5				
22795	SuperMP (PW)-H Guard column	4.6	3.5	8				
22796	SuperOligoPW Guard column	4.6	3.5	3				
08034	Oligo Guard column	6.0	4.0	13	For 7.8 mm ID G-Oligo-PW columns			
08033	PW$_{XL}$ Guard column	6.0	4.0	12	For 7.8 mm ID PW$_{XL}$& G-DNA-PW (TSKgel G3000PW packing)			
21876	PW$_{XL}$-CP Guard column	6.0	4.0	13	For 7.8 mm ID PW$_{XL}$-CP columns			
06763	PW-L Guard column	7.5	7.5	13	For 7.5 mm ID G1000PW & G2000PW (TSKgel G2000PW packing)			
06762	PW-H Guard column	7.5	7.5	13	For 7.5 mm ID G2500PW through GMPW columns			
06758	PW-H Guard column	21.5	7.5	17	For 21.5 mm ID G2500PW through G5000PW columns			
Bulk packing								
08035	PW$_{XL}$ Top-Off, 1 g wet resin			10	For all PW$_{XL}$ and G-DNA-PW columns			

TSKgel ALPHA AND SUPERAW GEL FILTRATION COLUMNS

Gel Filtration and Gel Permeation Chromatography of water-soluble and polar organic-soluble polymers

HIGHLIGHTS

- A unique hydrophilic, polyvinyl resin is available in conventional column dimensions (Alpha) and high throughput column format (SuperAW).
- Exhibits strong mechanical stability and minimal swelling characteristics
- A wide range of solvent compatibility, from 100% water to 100% non-polar organic solvents
- The reduced particle size and shorter column length of TSKgel SuperAW columns provide equivalent resolution in one half the time for high throughput applications.
- Unlike polystyrene-divinylbenzene (PS-DVB) resins that may adsorb polymers due to hydrophobic interaction, both the TSKgel Alpha and
- SuperAW columns allow for the separation of polymers soluble in methanol.
- Provide accurate molecular weight determination of samples in dimethyl formamide and exhibit normal retention of polystyrene polymers
- System peaks from salts in the eluent elute away from the oligomer of interest, providing accurate MW deter-minations.

COLUMN SELECTION

The **TSKgel Alpha Series** consists of six columns with three particle sizes: 7, 10, and 13 µm. These columns span a wide MW separation range from 100 to more than 1 x 10^6 Da when using polyethylene oxide (PEO) as a MW standard. Exclusion limits for the TSKgel Alpha columns for polyethylene oxide (PEO), polyethylene glycols (PEG) and polystyrenes (PS) are shown in the table below. Calibration curves for the TSKgel Alpha Series columns are shown on the next page for polyethylene oxide, polyethylene glycol and polystyrene standards.

The **TSKgel SuperAW series** contains a similar chemistry as the TSKgel Alpha series but offers the benefit of smaller particle sizes (4 µm to 9 µm) and smaller column dimensions. Reductions in analysis time and mobile phase consumption make SuperAW columns ideal for high throughput applications. TSKgel Alpha and SuperAW columns are offered in 5 discrete exclusion ranges and 1 mixed bed. Both column types can accommodate polymer standards up to several million Dalton molecular weight (see calibration curves on the next page

TABLE 3

Exclusion limits for TSKgel Alpha Series and SuperAW Series columns

TSKgel Column	Particle size (µm)	Exclusion limit (Da) for various standards and eluents		
		PEO[a]/H_2O	PS[b]/10 mmol/L LiBr in DMF	PEG[c]/10 mmol/L LiBr in MeOH
Alpha-2500	7	5 x 10^3	1 x 10^4	1 x 10^4
Alpha-3000	7	9 x 10^4	1 x 10^5	6 x 10^4
Alpha-4000	10	4 x 10^5	1 x 10^6	3 x 10^6
Alpha-5000	10	1 x 10^6	7 x 10^6	N.D.
Alpha-6000	13	> 1 x 10^7	> 1 x 10^7	N.D.
Alpha-M	13	> 1 x 10^7	> 1 x 10^7	N.D.
SuperAW2500	4	5 x 10^3	8 x 10^3	1 x 10^4
SuperAW3000	4	9 x 10^4	8 x 10^4	1 x 10^5
SuperAW4000	6	1 x 10^6	6 x 10^5	6 x 10^5
SuperAW5000	7	1 x 10^{6*}	N.D.	N.D.
SuperAW6000	9	1 x 10^{7*}	N.D.	N.D.
SuperAWM-H	9	1 x 10^{7*}	N.D.	N.D.

N.D. = not determined a Polyethylene oxide b Polystyrene divinyl benzene c Polyethylene glycol

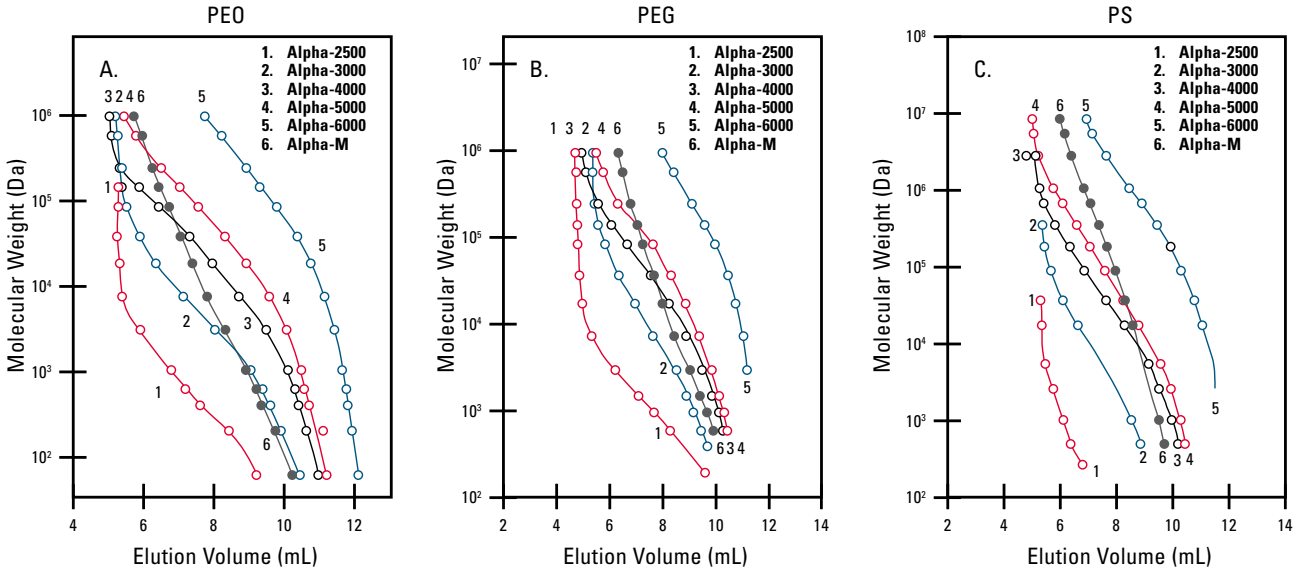
* Exclusion limit for SuperAW5000, SuperAW6000, and SuperAWM-H are estimated, respectively

SEC

CALIBRATION CURVES FOR TSKgel ALPHA AND SUPERAW GEL FILTRATION COLUMNS

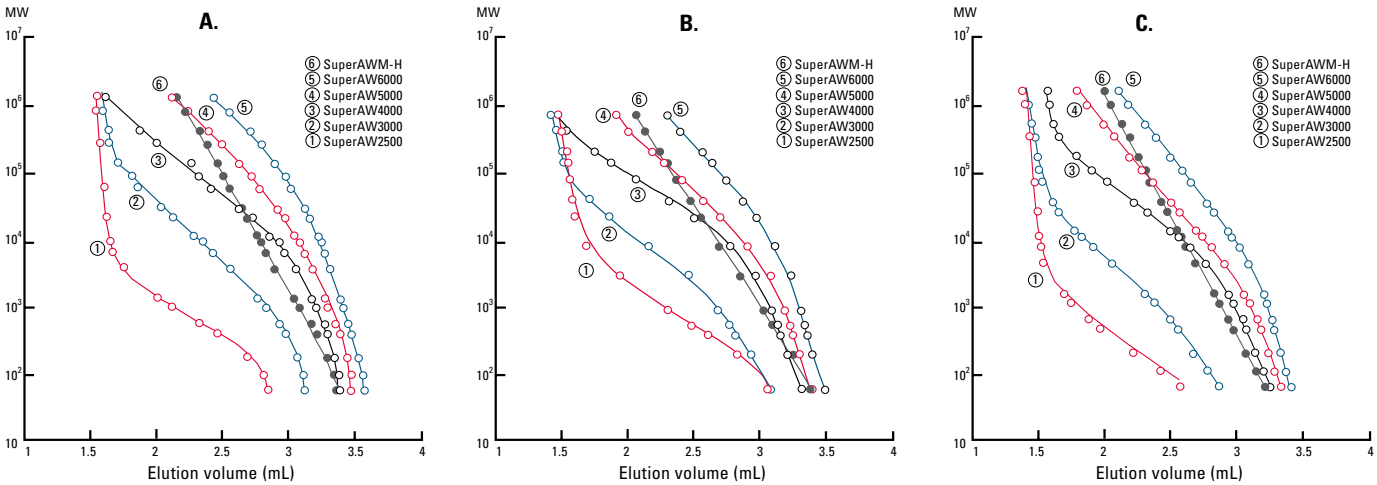
The best results are obtained when selecting a column with the sample's molecular weight in the linear portion of the calibration curve.

Polyethylene oxide (PEO), polyethylene glycol (PEG) and polystyrene (PS) calibration curves for TSKgel Alpha columns



Column: TSKgel Alpha Series, 7.8 mm ID x 30 cm L; Eluent: A. H_2O; B. 10 mmol/L LiBr in Methanol; C. 10 mmol/L LiBr in DMF; Flow Rate: 1.0 mL/min; Temperature: A. 25°C; B. 25°C; C. 40°C; Detection: RI

Calibration curves for TSKgel SuperAW series in different solvents with different polarity



Column: TSK-GEL SuperAW Series (6.0 mm ID x 15 cm L)

Eluent: A. Water; B. MeOH containing 10 mmol/L LiBr; C. DMF containing 10 mmol/L LiBr

Flow rate: 0.6 mL/min; Temperature: 25°C; Detection: Refractive index detector

Samples: Standard polyethylene oxide, polyethylene glycol, ethylene glycol

APPLICATIONS OF TSKgel ALPHA AND SUPERAW GEL FILTRATION COLUMNS

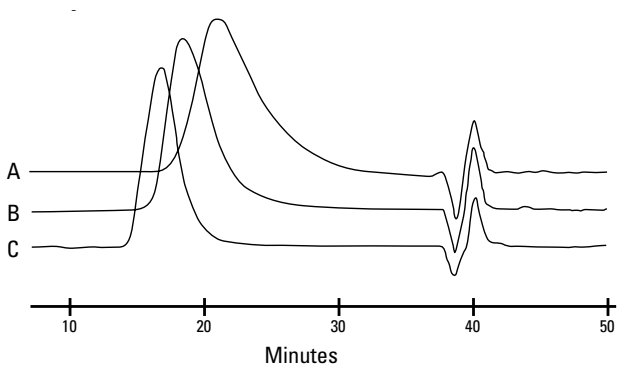
The versatility of using TSKgel Alpha columns with various polar solvents is illustrated in FIGURE 17 for the analysis of cellulose derivatives. A TSKgel Alpha-M column was used to separate ethylcellulose with the polar solvent DMF and ethylhydroxyethyl cellulose with methanol.

The separation of polyvinylalcohol with different degrees of saponification is shown in FIGURE 18. This separation was performed with a TSKgel Alpha-5000 and a TSKgel Alpha-3000 column in series using a hexafluoroisopropanol mobile phase.

FIGURE 19 shows that the column efficiency of TSKgel SuperAW series columns is maintained in a wide variety of polar organic solvents.

FIGURE 18

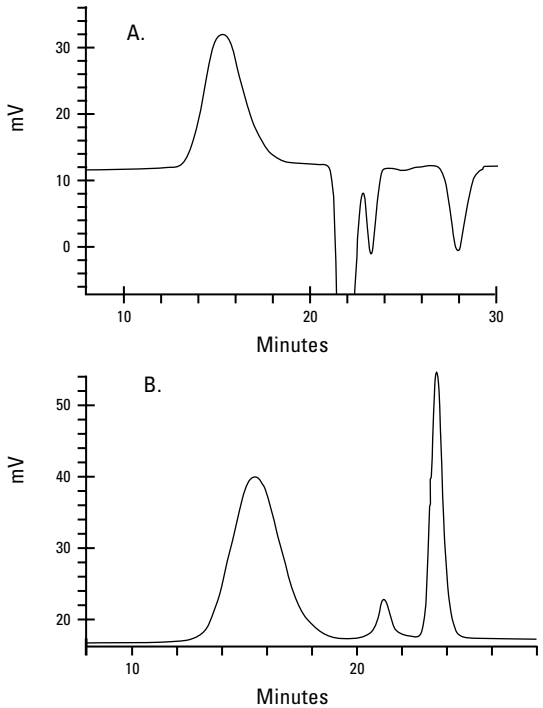
Polyvinylalcohol characterization using TSKgel Alpha-5000 and Alpha-3000 columns in series



Column: TSKgel Alpha-5000 and Alpha-3000, 7.8 mm ID x 30 cm L in series
Sample: degree of saponification of polyvinyl alcohol: A. 75%, B. 88%, C. 100%; Eluent: hexafluoroisopropanol (HFIP); Flow Rate: 0.5 mL/min; Temperature: 40°C; Detection: RI

FIGURE 17

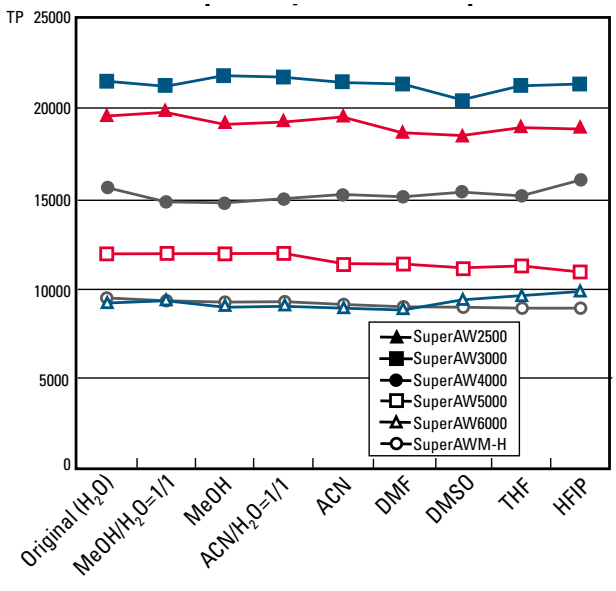
TSKgel Alpha-M separation of cellulose derivatives



Column: TSKgel Alpha-M, 7.8 mm ID x 30 cm L;
Sample: A. 50 µL ethylcellulose, 0.1%; B. 50 µL ethylhydroxyethylcellulose, 0.1%; Elution: A. 10 mmol/L LiBr in DMF; B. 10 mmol/L LiBr in methanol; Flow Rate: 0.5 mL/min; Temperature: 40°C; Detection: RI

FIGURE 19

Solvent Compatibility of TSKgel SuperAW series



Column: TSKgel SuperAW Series (6.0 mm ID x 15 cm L); Eluent: Water
Flow rate: 0.6 mL/min; Temperature: 25°C; Detection: Refractive index detector
Sample: Ethylene glycol; Inj. volume: 5 µL (2.5 g/L)

SEC

ORDERING INFORMATION

Part #	Description	ID (mm)	Length (cm)	Particle size (µm)	Number theoretical plates	Flow rate (mL/min) Range	Flow rate (mL/min) Max.	Maximum pressure drop (MPa)
TSKgel Stainless Steel Columns								
18339	Alpha-2500	7.8	30	7	≥ 16,000	0.5 - 0.8	1.0	4.0
18340	Alpha-3000	7.8	30	7	≥ 16,000	0.5 - 0.8	1.0	4.0
18341	Alpha-4000	7.8	30	10	≥ 10,000	0.3 - 0.6	1.0	3.0
18342	Alpha-5000	7.8	30	10	≥ 10,000	0.3 - 0.6	1.0	3.0
18343	Alpha-6000	7.8	30	13	≥ 7,000	0.3 - 0.6	1.0	2.0
18344	Alpha-M (mixed bed)	7.8	30	13	≥ 7,000	0.3 - 0.6	1.0	2.0
Guard columns								
18345	Alpha Guard column	6	4	13	For all Alpha columns			
TSKgel VMpak columns*								
20011	VMpak-25	2.0	5	7	≥ 1,000	0.1 - 0.2	0.25	20
20012	VMpak-25	2.0	15	7	≥ 3,000	0.1 - 0.2	0.25	60
TSKgel Stainless Steel Columns								
19315	SuperAW2500	6.0	15	4	≥ 16,000	0.3 - 0.6	0.6	60
19316	SuperAW3000	6.0	15	4	≥ 16,000	0.3 - 0.6	0.6	60
19317	SuperAW4000	6.0	15	6	≥ 10,000	0.3 - 0.6	0.6	40
19318	SuperAW5000	6.0	15	7	>10,000	0.3 - 0.6	0.6	30
19319	SuperAW6000	6.0	15	9	>7,000	0.3 - 0.6	0.6	20
19320	SuperAWM-H	6.0	15	9	>7,000	0.3 - 0.6	0.6	20
Guard columns								
19321	SuperAW-L Guard Column	4.6	3.5	7	For SuperAW2500-4000 columns.			
19322	SuperAW-H Guard Column	4.6	3.5	13	For SuperAW5000-AWM-H columns			

*TSKgel VMpak-25 series contains a similar packing as TSKgel Alpha-2500. It can be used for multimodal LC/LC-MS separations.

TSKgel HXL, HHR, SUPERH AND SUPERHZ GEL PERMEATION COLUMNS

Polymer-based columns for Gel Permeation Chromatography of organic-soluble polymers

HIGHLIGHTS

- Porous, highly cross-linked, spherical polystyrene divinylbenzene (PS-DVB) resin.
- Five different TSKgel H-type columns are available. Each of these are packed with different particle sizes (see table below).
- Expanded molecular weight ranges with exclusion limits from 1,000 Da to an estimated 4 x 10^8 Da
- Minimal shrinking and swelling of the column bed
- Chemically and thermally stable
- Use 4.6 & 6.0 mm ID SuperMultiporeHZ, SuperHZ and Super H columns for reduced solvent consumption in high throughput analysis.
- SuperMultiporeHZ and MultiporeHXL columns provide linear calibration curves over a wider MW range.
- Semi-micro SuperHZ columns now available as multipore columns with linear calibration curves.

TSKgel H Series columns are recommended for the analysis of organic-soluble polymers and are packed with spherical particles composed of polystyrene cross-linked with divinylbenzene (PS-DVB). Each line of columns within this series differs in degree of inertness and operating temperature range. The packings are available in eight pore sizes and span four different column chemistries. For polymer samples with a broad molecular range, packing of several pore sizes are provided in the mixed bed columns: TSKgel SuperHZM series, TSKgel SuperHM series, TSKgel GMHXL, TSKgel GMHHR, and selected high temperature versions provide linear calibration curves up to several million Daltons (see page 53).

COLUMN SELECTION

The Super prefix refers to the efficiency of the column. The Super series columns contain ultra efficient particles as small as 3 µm, housed in 15 cm length columns. The smaller particle allows for equivalent resolution to conventional HXL columns, with 50% less run time due to the shorter column length. The Super series columns are an excellent choice for high throughput polymer analysis.

TABLE 4

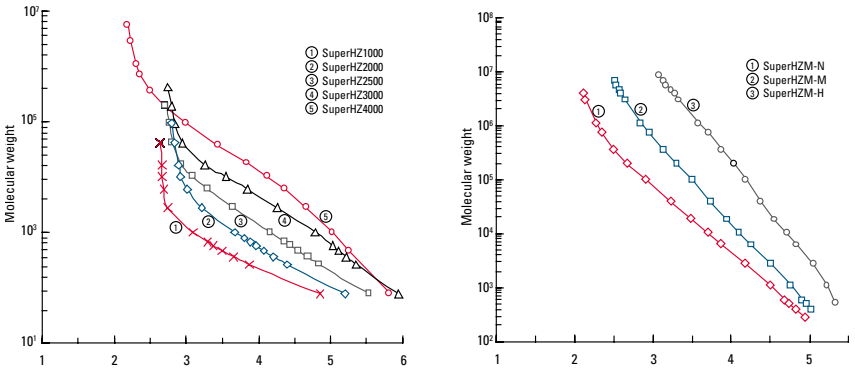
Series Type	SuperMultiporeHZ	SuperHZ	HXL	SuperH	HHR
Application focus	Ultra-high performance with a low dead volume and a wide pore distribution in each particle for superior linearity	High-throughput polymer analysis with ultra low polymer adsorption. Limited solvent compatibility range.	Conventional polymer analysis with ultra low polymer adsorption. Ltd solvent compatibility range.	High-throughput polymer analysis with expanded solvent compatibility.	Conventional polymer analysis with expanded solvent compatibility range.
Particle size	3, 4 and 6 µm, depending on pore size	3, 5 and 10 µm, depending on pore size	5 and 13 µm, depending on pore size	3 and 5 µm, depending on pore size	5 µm
Theoretical plates[1]		16,000/15 cm column	16,000/30 cm column	16,000/15 cm column	16,000/30 cm column
Maximum temperature		G1000 - G4000 60°C G5000 - mixed 80°C	G1000 - G4000 60°C G5000 - mixed 80°C	140°C	140°C
Standard shipping solvent	THF	THF	THF[2]	THF[2]	THF[2]
THF can be switched to	benzene, chloroform, toluene, xylene, dichloromethane[3] and dicholoroethane[3]			see our website for detailed information	
Other shipping solvents available?	yes[4]	yes[4]		no	
Number of solvent substitutions	One time only	One time only	One time only	Several[5]	Several[5]
Solvent exchange instructions	Linear gradient with a 2 %/min rate of change at a flow rate <0.25 mL/min.		Linear gradient with a 2 %/min rate of change at a flow rate <0.5 mL/min.	Linear gradient with a 2%/min rate of change according to flow rates listed on our website.	

1) Theoretical plates listed are based on smallest particle size listed
2) High-temperature columns (HT) are shipped with ODCB (Orthochlorodivinylbenzene) as standard shipping solvent.
3) Switching from THF to dichloromethane and dichloroethane is not recommended for G1000 pore size columns.
4) See our website for available shipping solvents
5) After switching to a very polar solvent such as acetone, switching back to a nonpolar solvent is not recommended.

SEC

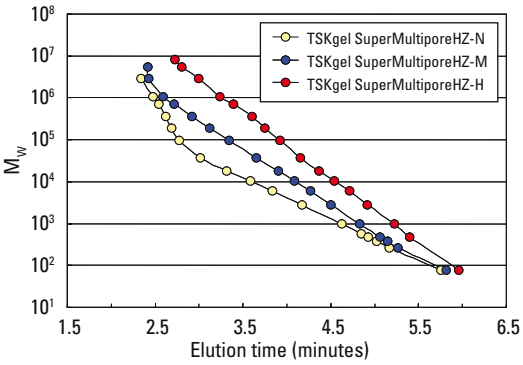
CALIBRATION CURVES FOR TSKgel H-TYPE GELPERMEATION COLUMNS

Calibration curves for TSKgel SuperHZ columns with polystyrene standards



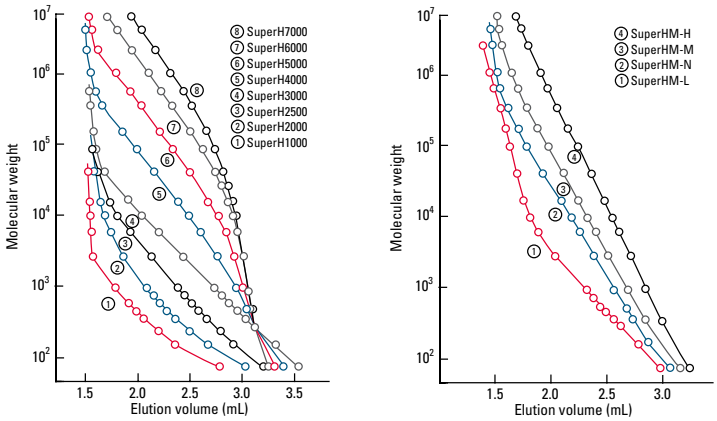
Column: TSKgel SuperHZ series (4.6 mm ID x 15 cm L); Eluent: THF; Flow rate: 0.35 mL/min; Temp.: 25°C; Sample: polystyrene standards; Inj. volume: 2 µL

Calibration curves for TSKgel SuperMultiporeHZ-M, H and N columns



Columns: TSKgel SuperMultiporeHZ-N, 3 µm, 4.6 mm ID x 15 cm L, TSKgel SuperMultiporeHZ-M, 4 µm, 4.6 mm ID x 15 cm L, TSKgel SuperMultiporeHZ-H, 6 µm, 4.6 mm ID x 15 cm L; Mobile phase: THF; Flow rate: 0.35 mL/min; Detection: UV@254nm; Temp.: 25°C; Samples: polystyrene standards

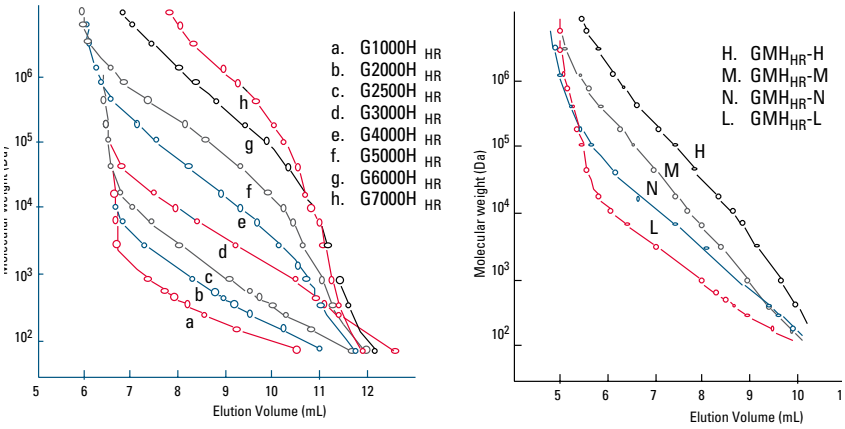
Calibration curves for TSKgel SuperH columns with polystyrene standards



Column: TSKgel SuperH series (6.0 mm ID x 15 cm L); Eluent: THF; Flow rate: 0.6 mL/min; Temp.: 25°C; Detection: UV@254 nm; Sample: polystyrene standards

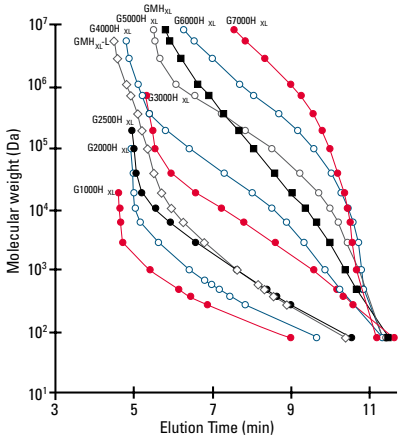
The best results are obtained when selecting a column with the sample's molecular weight in the linear portion of the calibration curve.

Calibration curves for TSKgel H_{HR} columns with polystyrene standards



Column: TSKgel H_{HR}series (7.8 mm ID x 30 cm L); Sample: polystyrene standards; Elution: THF Flow Rate: 1.0 mL/min; Temp.: 25°C; Detection: UV@254 nm

Calibration curves for TSKgel H_{XL} columns with polystyrene standards



Column size: 7.8 mm ID x 30 cm L; Sample: polystyrene standards; Eluent: THF; Flow Rate: 1.0 mL/min; Temp.: 25°C; Detection: UV @ 254 nm

MULTI-PORE SIZE DISTRIBUTION IN A POLYSTERENE PACKING MATERIAL

Novel approach to GPC of samples with a wide range of molecular weights

Prior to the introduction of TSKgel MultiporeHXL and SuperMultiporeHZ columns, scientists separating polymers with a wide range of molecular weights were left with two options. One option is to use multiple columns of different pore sizes linked together in series. A second is to use a column packed with a mixed bed resin of different pore sizes at an optimized mix ratio. However, problems can occur with both of these methods, which include distortion of the chromatogram or deviations between the actual calibration curve and the calibration curve approximated from data obtained from the molecular weight standards.

As is shown in Figure 20, a novel approach to solve this problem was developed by Tosoh scientists and is incorporated in TSKgel MultiporeHXL and SuperMultiporeHZ Series columns.

These columns are packed with particles of uniform size synthesized with a broad distribution of pore sizes. This novel approach creates a linear calibration curve within each particle. Therefore, columns with an extended linear calibration curve can now be prepared without mixing particles of different pore sizes. This results in sharper peaks without inflection points that may be observed using mixed-bed columns.

The pore size distributions of the TSKgel MultiporeHXL-M column and a mixed-bed column are shown in FIGURE 21. The mixed-bed column shows a sharp maximum for pores with a diameter of 0.08 µm, though the overall pore size distribution ranges from 0.006 to 0.6 µm in diameter. In the case of the TSKgel MultiporeHXL-M column, the pore size distribution exhibits a wider maximum range from 0.02 to 0.1 µm in diameter. This difference in pore size distribution may explain the reason for the inflection phenomenon.

The small ID (4.6 mm) and length (15 cm) of the SuperMultiporeHZ columns reduces solvent consumption and results in quick run times, and offers high throughput capabilities. FIGURE 22 demonstrates that inflection points are no longer observed with semi-micro columns packed from particles prepared by multi-pore technology.

FIGURE 20

Strategies for wide range separation using SEC

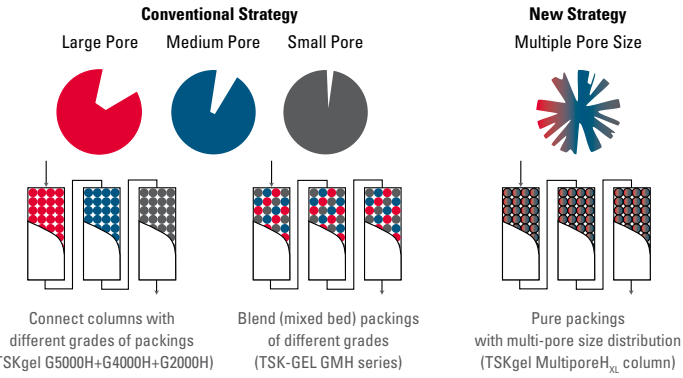


FIGURE 21

Pore size distribution of TSKgel MultiporeHXL-M column and a mixed-bed column

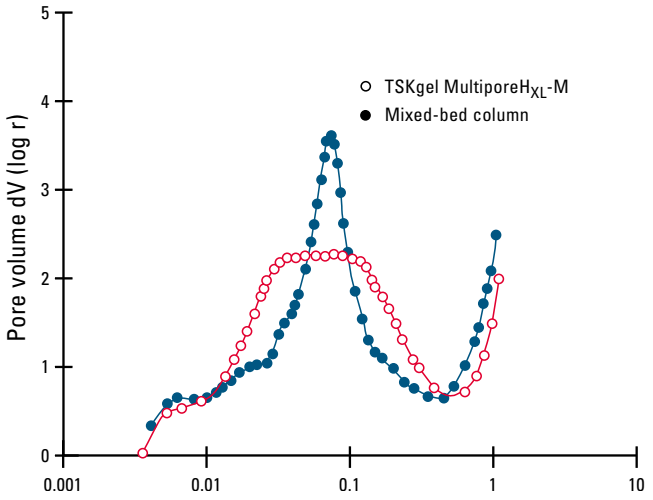
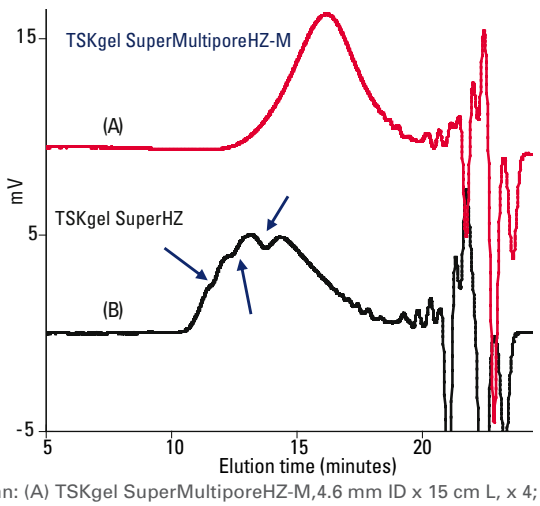


FIGURE 22

Comparison of TSKgel SuperMultiporeHZ-M and TSKgel SuperHZ for separation of Acrylic resin



Column: (A) TSKgel SuperMultiporeHZ-M,4.6 mm ID x 15 cm L, x 4;
(B) TSKgel SuperHZ4000+3000+2500+2000, 4.6 mm ID x 15 cm L x 4
Mobile phase: THF; Detection: RI; Temperature: 40°C; Injection vol.: 10 µL
Samples: acrylic resin

SEC

APPLICATIONS OF TSKgel H-TYPE GEL PERMEATION COLUMNS

PHTHALATE ESTERS

FIGURE 23 demonstrates the high efficiency separation on a TSKgel G1000HXL column for low molecular weight phthalate esters. Resolution was close to baseline, even though the molecular weights of the esters differed by less than 50 Da.

PHENOL RESIN

The TSKgel GMHXL-L column has been designed to provide a complete profile for high molecular weight samples that contain low molecular weight additives. The calibration curve for this mixed-bed column is shallow in the low molecular weight range of oligomers. Sample adsorption is not observed.

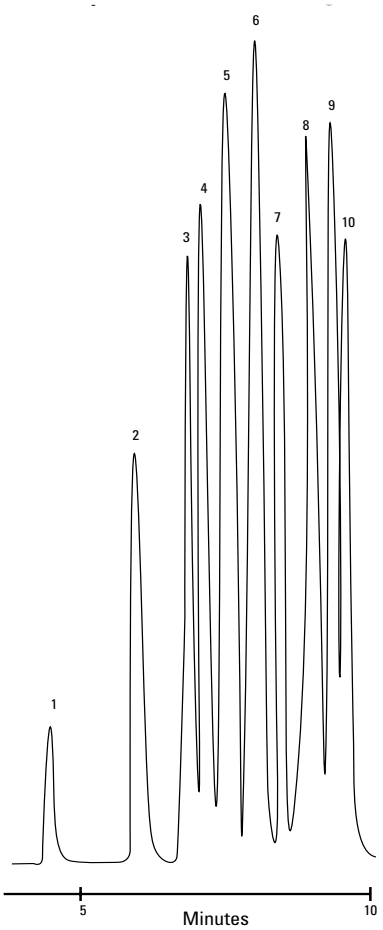
For example, the complete profile of a phenol resin, with high resolution of the low molecular weight components, is shown in FIGURE 24. Other applications for the TSKgel GMHXL-L column include analyses of paint materials, bond and adhesive components and synthetic polymer additives.

FATTY ACIDS

In FIGURE 25, two TSKgel G2000HXL columns in series separate a mixture of fatty acids ranging from C4 to C30.

FIGURE 23

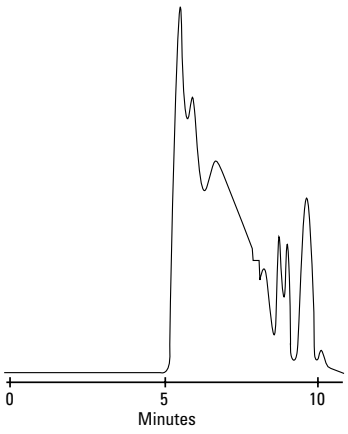
High resolution of phtalate ester on TSKgel G1000HXL



Column: TSKgel G1000HXL, 7.8 mm ID x 30 cm L;
Sample: 1. polystyrene (10,200Da), 2. dioctylphthalate (391Da), 3. dibutylphthalate (278Da), 4.dipropylphthalate (250Da), 5. diethylphthalate (222Da), 6. dimethylphthalate (194Da), 7. n-propylbenzene (120Da), 8. ethylbenzene (116Da), 9. toluene (92Da), 10. benzene (78Da); Elution: THF; Flow Rate: 1.0mL/min; Detection: UV@254nm

FIGURE 24

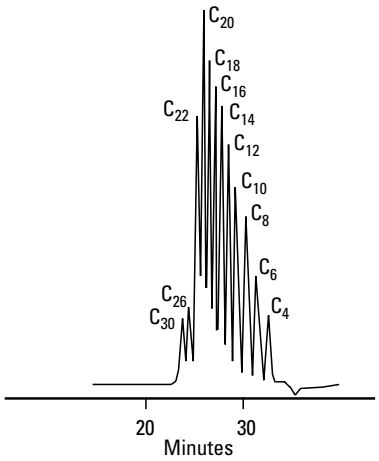
Separation of phenol resin on TSKgel GMHXL-L



Column: TSKgel GMHXL-L, 7.8 mm ID x 30 cm L;
Sample: phenol resin; Elution: THF; Flow Rate: 1.0 mL/min;
Detection: UV @ 254nm

FIGURE 25

Separation of fatty acid



Column: TSKgel G2000HXL, two 7.8 mm ID x 30 cm L in series;
Sample: fatty acids; Elution: THF; Flow Rate: 1.0 mL/min; Detection: RI

APPLICATIONS OF TSKgel H-TYPE GEL PERMEATION COLUMNS

ACRYLIC POLYMER

FIGURE 26 shows the separation of an acrylic polymer on the TSKgel MultiporeHXL-M column compared with two commercially available mixed-bed columns. The arrows illustrate the inflections seen in the chromatograms from mixed-bed columns and the improvement achieved when using the TSKgel MultiporeHXL-M column.

POLYMETHYLMETHACRYLATE

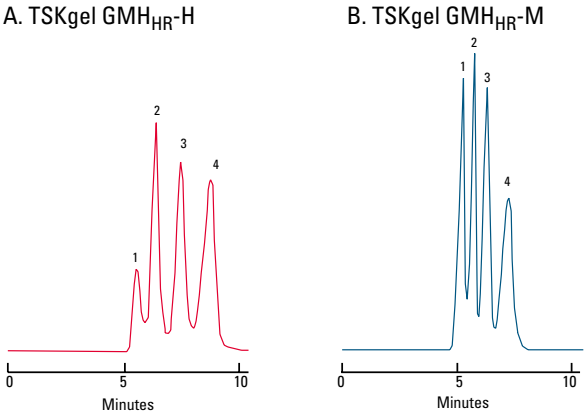
The effect of different pore size distributions in the mixed beds of TSKgel GMHHR-H and TSKgel GMHHR-M is illustrated in FIGURE 27. The TSKgel GMHHR-M produces better resolution in the 8 x 10^5 to 1 x 10^4 Da range.

SEMI-MICRO GPC

Semi-micro columns are referred to as such since their dimensions are smaller than conventional columns in terms of internal diameter as well as in length: 4.6 mm or 6 mm ID x 15 cm vs. 7.8 mm ID x 30 cm of conventional GPC columns. As shown in FIGURE 28, a TSKgel SuperMultiporeHZ-N column provides the same or higher resolution at a much shorter analysis time than multiple conventional sized columns linked together.

FIGURE 27

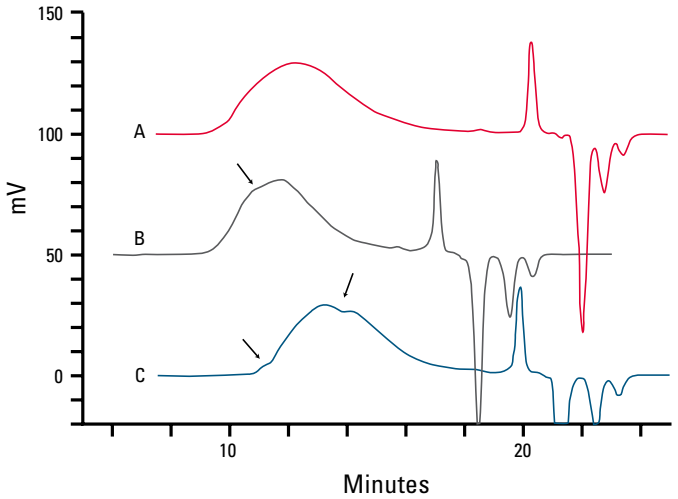
Comparison of TSKgel GMHHR-H and -M columns with polymethylmethacrylate standards



Columns: A. TSKgel GMHHR-H, 7.8 mm ID x 30 cm L;
B. TSKgel GMHHR-M, 7.8 mm ID x 30 cm L;
Sample: polymethylmethacrylate: 1. 820,000 Da, 2. 67,000 Da, 3. 10,200 Da, 4. 1,950 Da; Solvent: 5 mmol/L sodium trifluoroacetate in hexafluoroisopropanol; Flow Rate: 1.0 mL/min; Detection: UV@220 nm; Temperature: 40°C

FIGURE 26

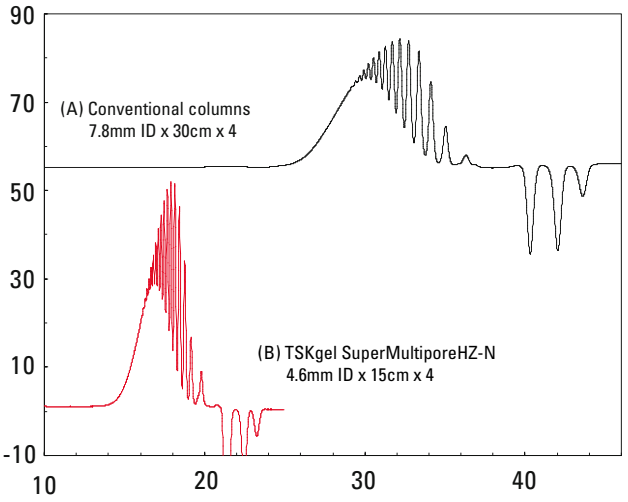
Separation of acrylic resin by SEC on TSKgel MultiporeHXL-M and mixed-bed type columns



Column: A. TSKgel MultiporeHXL-M, two 7.8 mm ID x 30 cm L in series,
B. Competitor P, two 7.5 mm ID x 30 cm L columns in series, mixed-bed type;
C. Competitor S, two 8.0 mm ID x 30 cm L columns in series, mixed-bed type;
Sample: acrylic polymer (0.1%, 50 µL); Elution: THF; Flow Rate: 1.0 mL/min; Temperature: 40°C; Detection: RI

FIGURE 28

PTMEG Analysis on Conventional and semi-micro TSKgel Columns



Columns: A. Conventional columns, 7.8 mm ID x 30 cm L x 4, B. TSKgel SuperMultiporeHZ-N, 4.6 mm ID x 15 cm L x 4;
Mobile phase: THF; Flow rate: (A) 1.0 mL/min (B) 0.35 mL/min; Temperature: 40°C; Injection vol.: (A) 60 µL (B) 10 µL; Sample: poly(teramethylene ether glycol), (PTMEG 650), 10 µg/µL

SEC

ORDERING INFORMATION

Part #	Description	ID (mm)	Length (cm)	Particle size (µm)	Number theoretical plates	Flow rate (mL/min) Range	Flow rate (mL/min) Max.	Maximum pressure drop (MPa)
TSKgel Stainless Steel Columns								
17352	G1000H$_{HR}$	7.8	30	5	≥ 16,000	0.5 - 1.0	2.0	5.0
17353	G2000H$_{HR}$	7.8	30	5	≥ 16,000	0.5 - 1.0	2.0	5.0
17354	G2500H$_{HR}$	7.8	30	5	≥ 16,000	0.5 - 1.0	2.0	5.0
17355	G3000H$_{HR}$	7.8	30	5	≥ 16,000	0.5 - 1.0	2.0	5.0
17356	G4000H$_{HR}$	7.8	30	5	≥ 16,000	0.5 - 1.0	2.0	5.0
17357	G5000H$_{HR}$	7.8	30	5	≥ 16,000	0.5 - 1.0	2.0	5.0
17358	G6000H$_{HR}$	7.8	30	5	≥ 16,000	0.5 - 1.0	2.0	5.0
17359	G7000H$_{HR}$	7.8	30	5	≥ 16,000	0.5 - 1.0	2.0	5.0
17362	GMH$_{HR}$-L mixed-bed	7.8	30	5	≥ 16,000	0.5 - 1.0	2.0	5.0
18055	GMH$_{HR}$-N mixed-bed	7.8	30	5	≥ 16,000	0.5 - 1.0	2.0	5.0
17392	GMH$_{HR}$-M mixed-bed	7.8	30	5	≥ 16,000	0.5 - 1.0	2.0	5.0
17360	GMH$_{HR}$-H mixed-bed	7.8	30	5	≥ 16,000	0.5 - 1.0	2.0	5.0
18393	GMH$_{HR}$-H(S)HT mixed-bed	7.8	30	13	≥ 8,000	5 - 1.0	2.5	2.0
18391	GMH$_{HR}$-H(30)HT mixed-bed	7.8	30	30	≥ 4,000			
18392	GMH$_{HR}$-H(20)HT mixed-bed	7.8	30	20	≥ 6,000			
16131	G1000H$_{XL}$	7.8	30	5	≥ 16,000	0.5 - 1.0	1.0	5.0
16134	G2000H$_{XL}$	7.8	30	5	≥ 16,000	0.5 - 1.0	1.2	5.0
16135	G2500H$_{XL}$	7.8	30	5	≥ 16,000	0.5 - 1.0	1.2	5.0
16136	G3000H$_{XL}$	7.8	30	5	≥ 16,000	0.5 - 1.0	1.2	3.5
16137	G4000H$_{XL}$	7.8	30	5	≥ 16,000	0.5 - 1.0	1.2	3.5
16138	G5000H$_{XL}$	7.8	30	9	≥ 14,000	0.5 - 1.0	1.2	1.5
16139	G6000H$_{XL}$	7.8	30	9	≥ 14,000	0.5 - 1.0	1.2	1.5
16140	G7000H$_{XL}$	7.8	30	9	≥ 14,000	0.5 - 1.0	1.2	1.5
16141	GMH$_{XL}$ mixed-bed	7.8	30	9	≥ 16,000	0.5 - 1.0	1.2	1.5
07112	GMH$_{XL}$-HT	7.8	30	13	≥ 5,500	5 - 1.0	1.2	1.5
16652	GMH$_{XL}$-L mixed-bed	7.8	30	5	≥ 16,000	0.5 - 1.0	1.2	3.5
18403	Multipore H$_{XL}$-M	7.8	30	5	≥ 16,000	0.5 - 1.0	1.0	3.5
17990	SuperH1000	6.0	15	3	≥ 16,000	0.3 - 0.6	0.8	6.0
17991	SuperH2000	6.0	15	3	≥ 16,000	0.3 - 0.6	0.8	6.0
17992	SuperH2500	6.0	15	3	≥ 16,000	0.3 - 0.6	0.8	6.0
17993	SuperH3000	6.0	15	3	≥ 16,000	0.3 - 0.6	0.8	4.0
17994	SuperH4000	6.0	15	3	≥ 16,000	0.3 - 0.6	0.8	4.0
17995	SuperH5000	6.0	15	3	≥ 16,000	0.3 - 0.6	0.8	4.0
17996	SuperH6000	6.0	15	5	≥ 16,000	0.3 - 0.6	0.8	4.0
17997	SuperH7000	6.0	15	5	≥ 16,000	0.3 - 0.6	0.8	4.0
17998	SuperHM-L	6.0	15	3	≥ 16,000	0.3 - 0.6	0.8	4.0
17999	SuperHM-N	6.0	15	3	≥ 16,000	0.3 - 0.6	0.8	4.0
18000	SuperHM-M	6.0	15	3	≥ 16,000	0.3 - 0.6	0.8	4.0
18001	SuperHM-H	6.0	15	3	≥ 16,000	0.3 - 0.6	0.8	4.0

ORDERING INFORMATION

Part #	Description	ID (mm)	Length (cm)	Particle size (µm)	Number theoretical plates	Flow rate (mL/min) Range	Flow rate (mL/min) Max.	Maximum pressure drop (MPa)
TSKgel Stainless Steel Columns								
19309	TSKgel SuperHZ1000	4.6	15	3	≥ 16,000	0.15 - 0.35	0.4	5.6
19302	TSKgel SuperHZ1000	6.0	15	3	≥ 16,000	0.25 - 0.6	0.7	5.6
19310	TSKgel SuperHZ2000	4.6	15	3	≥ 16,000	0.15 - 0.35	0.4	5.0
19303	TSKgel SuperHZ2000	6.0	15	3	≥ 16,000	0.25 - 0.6	0.7	5.0
19311	TSKgel SuperHZ2500	4.6	15	3	≥ 16,000	0.15 - 0.35	0.4	4.0
19304	TSKgel SuperHZ2500	6.0	15	3	≥ 16,000	0.25 - 0.6	0.7	4.0
19312	TSKgel SuperHZ3000	4.6	15	3	≥ 16,000	0.15 - 0.35	0.4	3.0
19305	TSKgel SuperHZ3000	6.0	15	3	≥ 16,000	0.25 - 0.6	0.7	3.0
19313	TSKgel SuperHZ4000	4.6	15	3	≥ 16,000	0.15 - 0.35	0.4	3.5
19306	TSKgel SuperHZ4000	6.0	15	3	≥ 16,000	0.25 - 0.6	0.7	3.5
19660	TSKgel SuperHZM-N	4.6	15	3	≥ 16,000	0.15 - 0.35	0.4	3.5
19661	TSKgel SuperHZM-N	6.0	15	3	≥ 16,000	0.25 - 0.6	0.7	3.5
19662	TSKgel SuperHZM-M	4.6	15	3 and 5	≥ 16,000	0.15 - 0.35	0.4	2.0
19663	TSKgel SuperHZM-M	6.0	15	3 and 5	≥ 16,000	0.25 - 0.6	0.7	2.0
19664	TSKgel SuperHZM-H	4.6	15	10	≥ 9,000	0.15 - 0.35	0.4	1.0
19665	TSKgel SuperHZM-H	6.0	15	10	≥ 9,000	0.25 - 0.6	0.7	1.0
21488	SuperMultiporeHZ-M	4.6	15	4	≥ 16,000			2.4
21815	SuperMultiporeHZ-N	4.6	15	3	≥ 20,000			4.0
21885	SuperMultiporeHZ-H	4.6	15	6	≥ 11,000			1.0

Guard columns					
18404	MultiporeH_{XL}-M Guard	6.0	4.0	5	For P/N 18403
07113	H_{XL} Guard Column	6.0	4.0		For G1000H_{XL} through G4000H_{XL} columns
13727	H_{XL} Guard Column	6.0	4.0		For G5000H_{XL} through GMH_{XL}-L mixed-bed columns
17368	H_{HR} Guard Column	6.0	4.0	5	For G1000-4000H_{HR} and GMHhr-L columns
17369	H_{HR} Guard Column	6.0	4.0	5	For G5000-7000H_{HR} and and GMH_{HR}-M; -N; -H columns
18002	SuperH Guard Column	4.6	3.5	3	For SuperH1000-4000
18003	SuperH Guard Column	4.6	3.5	3	For SuperH5000-7000 and HM-L;-N;-M;-H columns
18004	SuperH-RC Ref. Column	6.0	15		
19314	SuperHZ Guard Column	4.6	2.0	3	For 4.6 mm ID SuperHZ1000-4000 and HZM-N &-M columns
19668	SuperHZ Guard Column	4.6	2.0	10	For 4.6 mm ID SuperHZM-H columns
19666	SuperHZ Guard Column	4.6	3.5	3	For 6.0 mm ID SuperHZ1000-4000 and HZM-N &-M columns
19667	SuperHZ Guard Column	4.6	3.5	10	For 6.0 mm ID SuperHZM-H columns
21489	SuperMP-M Guard	4.6	2.0	4	For SuperMultipore HZ-M P/N 21488
21816	SuperMP-N Guard	4.6	2.0	3	For SuperMultipore HZ-N P/N 21815
21886	SuperMP-H Guard	4.6	2.0	6	For SuperMultipore HZ-H P/N 21887

SEC

ECOSEC GPC SYSTEM - BASED ON 35 YEARS OF EXPERIENCE IN GPC

EcoSEC is a compact, all-in-one GPC system for fast, high resolution, semi-micro GPC. Comprising a precision solvent delivery system, automatic injector, column oven and a high performance refractive index detector, the design of the system components, their configuration and the optimized flow line provides outstanding performance with minimized dead volume. This makes EcoSEC the ideal instrument to be used in combination with the well respected TSKgel semi-micro GPC/SEC columns. In Europe, EcoSEC is offered in cooperation with Polymer Standards Service (PSS), an acknowledged leader in the field of polymer analysis.