

# [ Size Exclusion Chromatography— GPC & GFC ]

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## Introduction



The Gel Permeation Chromatography (GPC) technique, pioneered by J.C. Moore of the Dow Chemical Company<sup>1</sup> in the early 1960s, is a mode of HPLC for separating molecules based upon their effective size in solution.

Originally patented and licensed to Waters as a method for characterizing polymers, GPC is also useful for separating small molecules from interfering matrices such as those found in foods, pharmaceutical preparations, and natural products. In addition, GPC is often used as the first step in the sequential analysis of complex, unknown, organic mixtures. In these analyses, the unknown mixture is separated by size in the GPC mode, purified fractions are collected, and the fractions are subsequently separated based on chemical differences by normal, or reversed-phase chromatography.

### Small Molecules in Organic or Aqueous Solution

Small molecules (less than 2,000 MW) can generally be separated if their size in solution differs by approximately 10%. GPC can also easily detect the presence of high molecular weight components in a sample matrix—indicating the need for sample cleanup prior to analysis of small molecules by normal, or reversed-phase chromatography.

### GPC of Organic Soluble Polymers

GPC is a practical, easy, and convenient tool for determining the complete molecular weight distribution of a polymer. Any oligomers, monomers and additives in a complex polymeric solution can also be separated as long as there exist significant size differences among these components.

GPC is an officially recognized method in the plastics industry and has been adopted by the American Society for Testing and Materials (ASTM, Committee D-20) for determining the molecular weight distribution of polymers.<sup>2</sup>

### Gel Filtration of Water Soluble Polymers and Proteins

Water soluble polymers and proteins can be routinely separated by size in solution. Size separations in aqueous solutions, often called gel filtration, are commonly used as the first step in the analysis of a complex protein mixture and as an important tool to characterize the molecular weights of water soluble polymers such as polyacrylamide.

### Selecting a Column

For over 40 years, Waters has been the market leader in GPC analysis, providing the highest quality GPC products and expert applications support. To solve unique separation problems across a broad spectrum of applications, we manufacture a wide range of GPC columns and accessories. Our series of Ultrastyrigel™ and μStyrigel™ columns have been the standard in GPC columns for the separation of organic-soluble samples. To solve your specific problems in aqueous separations, you can select from our Ultrahydrogel columns.

### Calibration Standards

Waters offers calibration standards for organic and aqueous GPC/SEC. Standards are available as individual molecular weights, or come in kits containing a range of MW standards.

### Waters Styragel Columns for Polymer Characterization

Designed specifically for polymer characterization, the Styragel® columns are grouped into the HR series for low-to-mid molecular weight samples, the HT series for high-temperature applications, and the HMW series for ultra-high molecular weight samples. Specially controlled styrene divinylbenzene formulations provide reproducible performance in your GPC applications.

<sup>1</sup> J.C. Moore, J. Polymer Science, A2, 835 (1964)

<sup>2</sup> ASTM D3536-76, D3593-80 and D3016-78

## Waters Styragel HR Columns

The Styragel HR (high resolution) series of columns were specifically developed for the analysis of low-to-mid molecular weight samples. The columns are packed with rigid 5  $\mu\text{m}$  styrene divinylbenzene particles and deliver the maximum resolution and efficiency required for low molecular weight analysis.

## Waters Styragel HT Columns

The Styragel HT (high temperature) series of columns were specifically developed for use in the mid-to-high molecular weight range. The columns are packed with rigid 10  $\mu\text{m}$  styrene divinylbenzene particles and can be used at ambient or high temperature while still maintaining excellent resolution. The narrow particle size distribution results in a more stable packed bed structure making Styragel HT columns extremely durable.

## Waters Styragel HMW Columns

The Styragel HMW (high molecular weight) series of columns were specifically developed for the analysis of shear sensitive, ultra-high molecular weight polymers. Styragel HMW columns are packed with rigid 20 micron styrene divinylbenzene particles and fitted with specially designed high porosity frits which minimize polymer shear effects. The columns may be used at either ambient or elevated temperature and exhibit excellent column lifetime.

Your choice of conventional 7.8 mm i.d. or solvent-efficient 4.6 mm i.d. columns. In addition to the conventional 7.8 mm columns, the three Styragel series include 4.6 mm i.d. columns in both the single-pore and mixed-bed columns. These Styragel narrow-bore columns can cut your solvent consumption and disposal costs by as much as two thirds. When used with low-dispersion GPC systems, our 4.6 mm columns match the high-performance of our 7.8 mm columns.

We also carry a full range of high resolution aqueous GPC columns, preparative GPC columns and calibration standards.

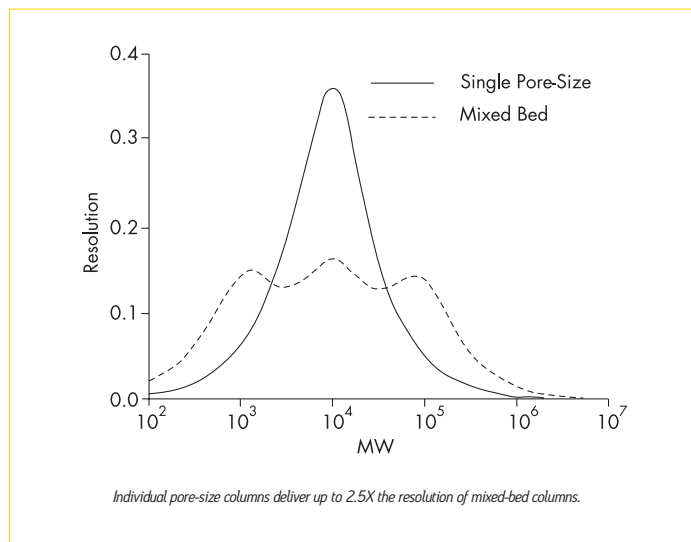
## Styragel Guard Column

The Styragel 4.6 mm i.d. x 30 mm guard column is designed to increase the lifetime of your Styragel analytical column. The guard column can be used in series with any Waters conventional organic GPC column.

## Column Bank Optimization

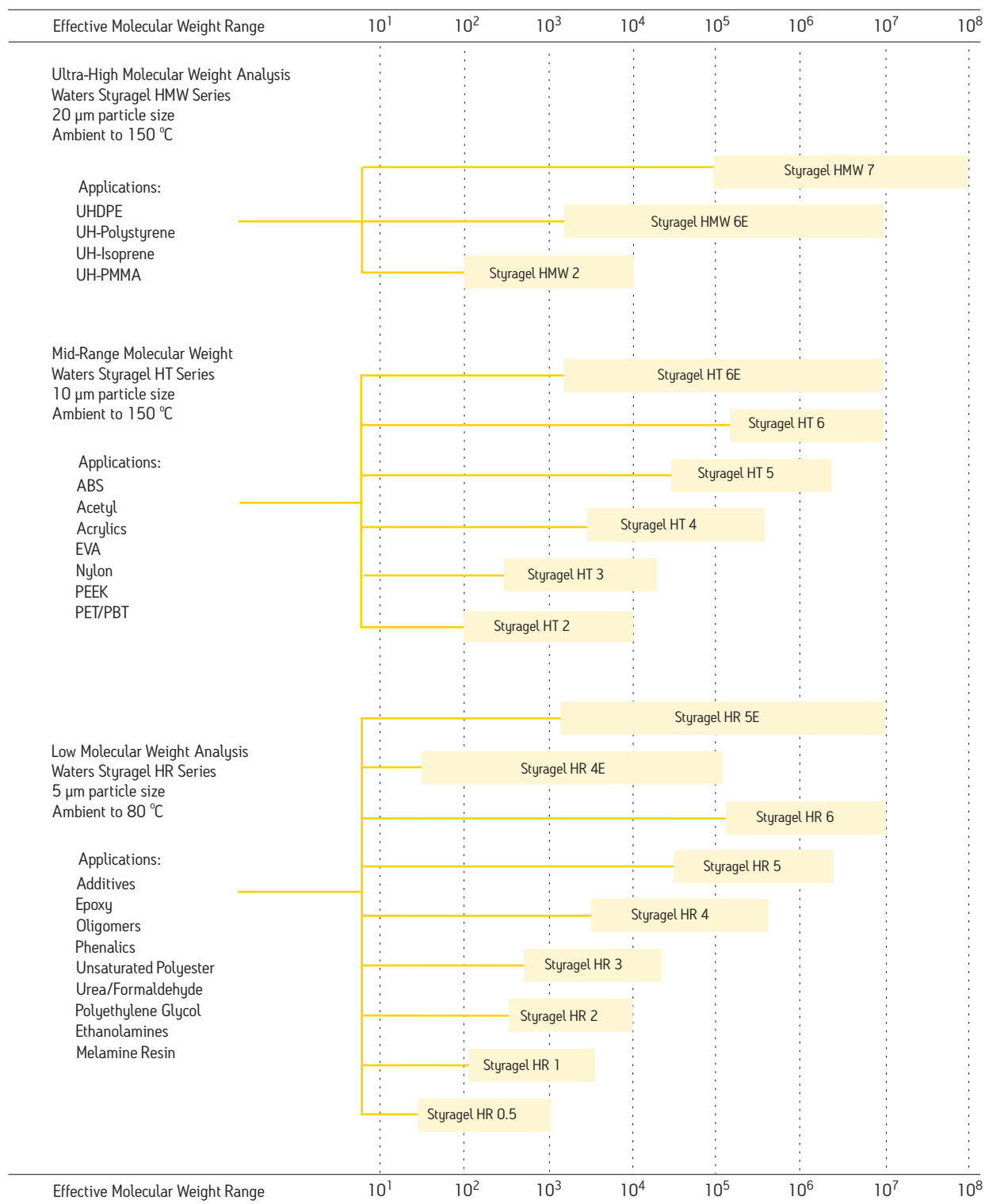
Using the proper column is essential to optimum performance. The rule for selecting the best column(s) for an analysis is straightforward—provide separation only for the molecules which you wish to separate. Never specify a column with a higher exclusion limit than the exclusion limit required by the largest molecules you wish to separate. When the measurement of broad distributions is desired, mixed bed or extended range columns are appropriate, thereby resulting in separation power that is constant at all molecular weight sizes.

Waters Styragel column offering is comprised of mixed-bed and narrow molecular weight range columns. The mixed-bed columns, designated “E” for extended range, are ideal for use as scouting columns when the molecular weight range of your sample is unknown or for the measurement of samples with broad distributions. The narrow molecular weight range columns deliver greater pore volume and resolution in a more concentrated molecular weight range and are a much more powerful tool for obtaining more precise molecular weight information.

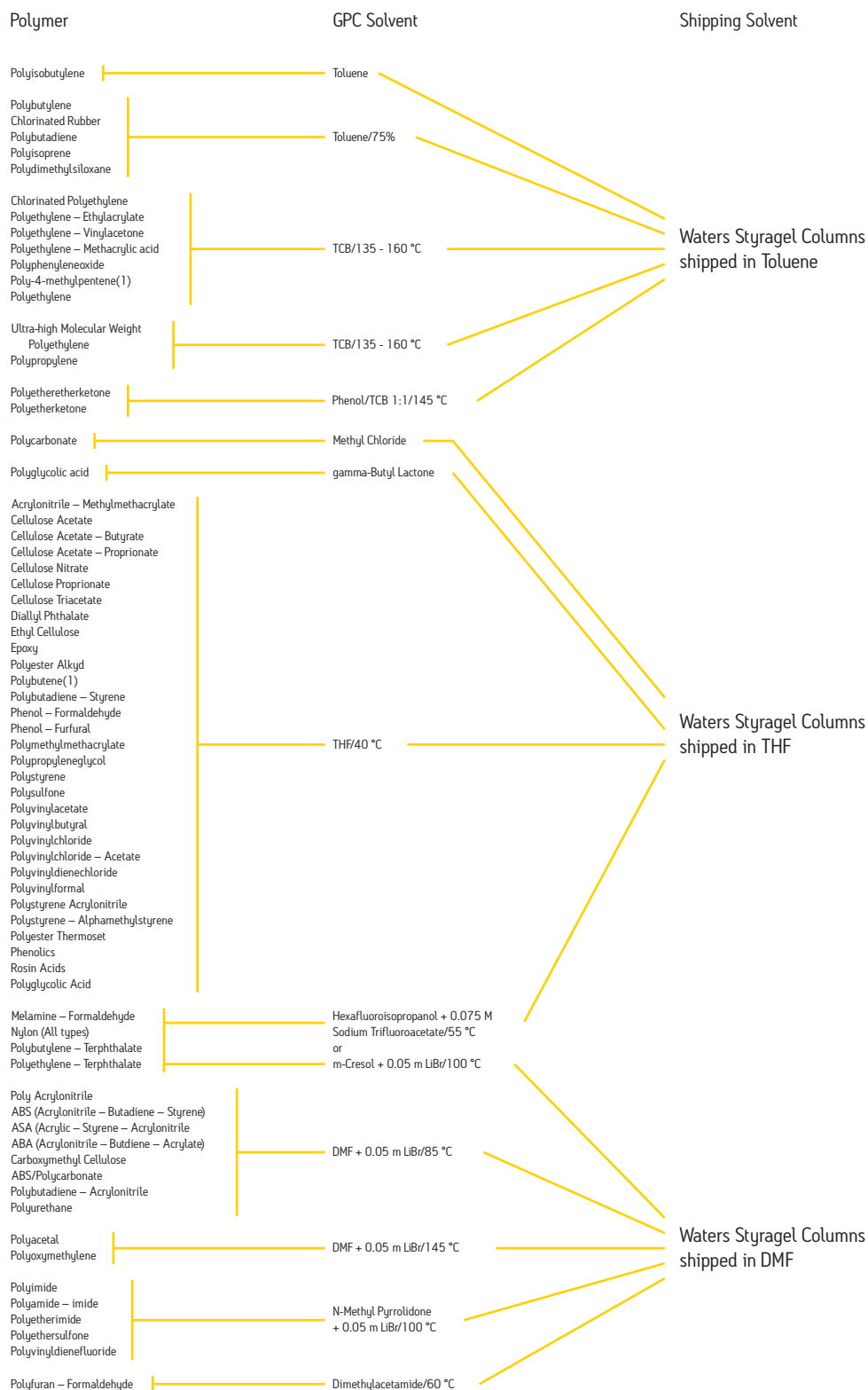




# GPC Column Selection Guide



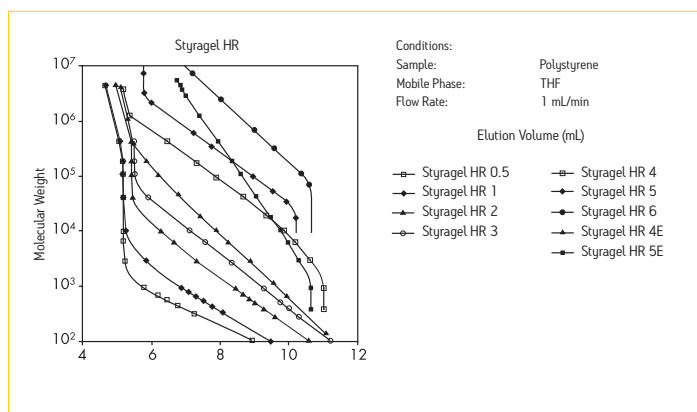
# GPC Solvent Selection Guide



## Styragel HR High-Resolution Columns

Designed particularly for low molecular weight samples, the Waters Styragel HR (high resolution) columns are ideal for the analysis of oligomers, epoxies, and polymer additives where high resolution is critical. Packed with rigid 5  $\mu\text{m}$  particles, these columns deliver unrivaled resolution and efficiency in the low-to-mid molecular weight region.

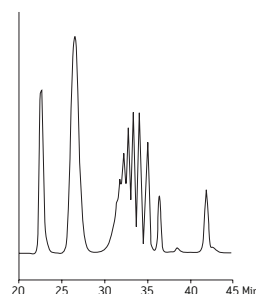
### Calibration Curves for the Waters Styragel HR Series of High-Resolution Columns



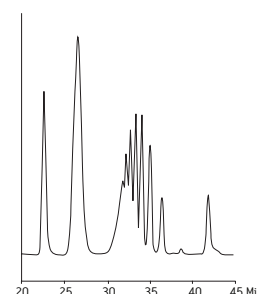
### Styragel HR High Resolution Columns for Unrivaled Resolution of Low Molecular Weight Samples

Column Bank:	Styragel HR 0.5, 1, 2, and 3	Column Bank:	Styragel HR 0.5, 1, 2, and 3
Mobile Phase:	THF	Mobile Phase:	THF
Temperature:	35 °C	Temperature:	35 °C
Flow Rate:	1 mL/min	Flow Rate:	0.35 mL/min
Sample:	Polystyrene standards: 0.5K, 5.05K, 49.8K	Sample:	Polystyrene standard mix 0.5K, 5.05K, 49.8K

Conventional Styragel HR Columns  
7.8 x 300 mm



Solvent-Efficient Styragel HR Columns  
4.6 x 300 mm



The 4.6 x 300 mm solvent-efficient Styragel columns deliver the same high resolution as our conventional 7.8 x 300 mm Styragel columns, with the added advantage of reducing solvent consumption by two-thirds.

### Styragel HR Columns (7.8 mm i.d. x 300 mm)

Effective Molecular Column	Part No. Weight Range	Part No. THF	Part No. DMF	Toluene
Styragel HR 0.5	0-1,000	WAT044231	WAT044232	WAT044230
Styragel HR 1	100-5,000	WAT044234	WAT044235	WAT044233
Styragel HR 2	500-20,000	WAT044237	WAT044238	WAT044236
Styragel HR 3	500-30,000	WAT044222	WAT044223	WAT044221
Styragel HR 4	5,000-600,000	WAT044225	WAT044226	WAT044224
Styragel HR 4E	50-100,000	WAT044240	WAT044241	WAT044239
Styragel HR 5	50,000-4 x 10 <sup>6</sup>	WAT054460	WAT054466	WAT054464
Styragel HR 5E	2,000-4 x 10 <sup>6</sup>	WAT044228	WAT044229	WAT044227
Styragel HR 6	200,000-1 x 10 <sup>7</sup>	WAT054468	WAT054474	WAT054470
Styragel Guard Column 4.6 x 30 mm		WAT054405	WAT054415	WAT054410

### Styragel HR Columns (4.6 mm i.d. x 300 mm)

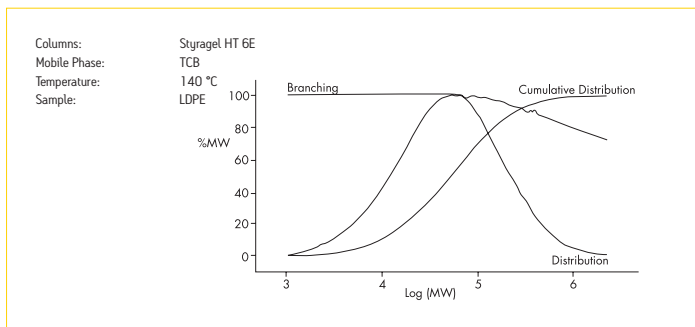
The same high performance as our conventional Styragel HR columns\* with the added advantage of reducing your solvent consumption by two-thirds.

Effective Molecular Column	Part No. Weight Range	Part No. THF	Part No. DMF	Toluene
Styragel HR 0.5	0-1,000	WAT045835	WAT045840	WAT045830
Styragel HR 1	100-5,000	WAT045850	WAT045855	WAT045845
Styragel HR 2	500-20,000	WAT045865	WAT045870	WAT045860
Styragel HR 3	500-30,000	WAT045880	WAT045885	WAT045875
Styragel HR 4	5,000-600,000	WAT045895	WAT045900	WAT045890
Styragel HR 4E	50-100,000	WAT045805	WAT045810	WAT045800
Styragel HR 5E	2,000-4 x 10 <sup>6</sup>	WAT045820	WAT045825	WAT045815

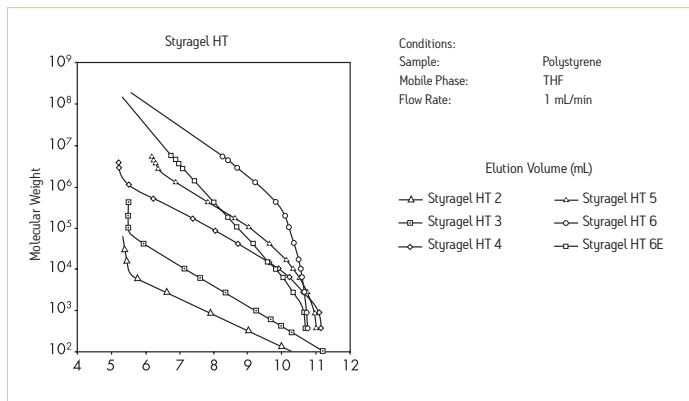
## Styragel HT High-Temperature, High-Stability Columns

The Waters Styragel HT (high temperature) columns can be used with aggressive solvents at high temperatures without sacrificing resolution or column lifetime. Packed with rigid 10  $\mu\text{m}$  particles, they have a typical plate count greater than 10,000 plates per column, these columns are extremely durable due to a narrow particle size distribution that results in a very stable column bed. Suitable for both ambient and high-temperature analysis, the Styragel HT columns offer excellent resolution of polymers in the mid-to-high molecular weight range.

### Styragel HT Columns Deliver Superior Performance — Even at High Temperatures



### Calibration Curves for the Waters Styragel HT Series of High-Temperature Columns



### Styragel HT Columns (7.8 mm i.d. x 300 mm)

Effective Molecular Column	Part No. Weight Range	Part No. THF	Part No. DMF	Part No. Toluene
Styragel HT 2	100-10,000	WAT054475	WAT054480	WAT054476
Styragel HT 3	500-30,000	WAT044207	WAT044208	WAT044206
Styragel HT 4	5,000-600,000	WAT044210	WAT044211	WAT044209
Styragel HT 5	50,000-4 x 10 <sup>6</sup>	WAT044213	WAT044214	WAT044212
Styragel HT 6	200,000-1 x 10 <sup>7</sup>	WAT044216	WAT044217	WAT044215
Styragel HT 6E	5,000 - 1 x 10 <sup>7</sup>	WAT044219	WAT044220	WAT044218
Styragel Guard Column 4.6 x 30 mm		WAT054405	WAT054415	WAT054410

### Styragel HT Columns (4.6 mm i.d. x 300 mm)

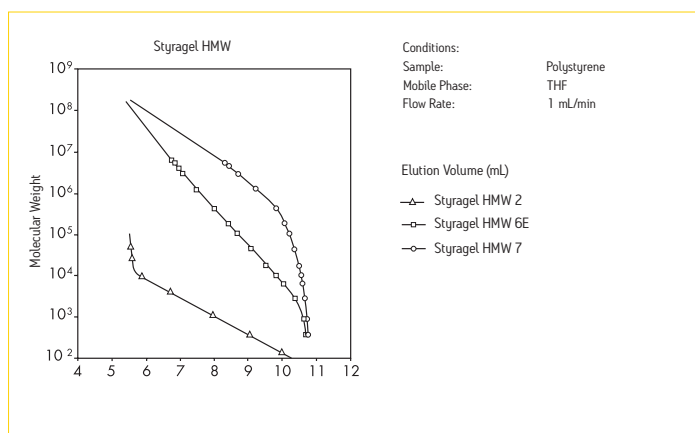
The same high performance as our conventional Styragel HT columns with the added advantage of reducing your solvent consumption by two-thirds.

Effective Molecular Column	Part No. Weight Range	Part No. THF	Part No. DMF	Part No. Toluene
Styragel HT 3	500-30,000	WAT045920	WAT045925	WAT045915
Styragel HT 4	5,000-600,000	WAT045935	WAT045940	WAT045930
Styragel HT 5	50,000-4 x 10 <sup>6</sup>	WAT045950	WAT045955	WAT045945
Styragel HT 6	200,000-1 x 10 <sup>7</sup>	WAT045965	WAT045970	WAT045960
Styragel HT 6E	5,000-1 x 10 <sup>7</sup>	WAT045980	WAT045985	WAT045975

## Styragel HMW High Molecular Weight Columns

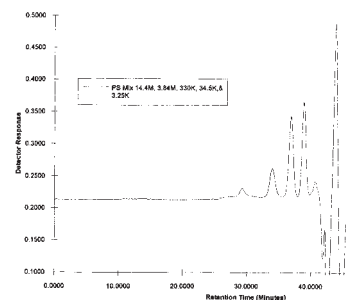
The Styragel HMW (high molecular weight) columns were specifically designed for analysis of ultra-high molecular weight polymers susceptible to shearing. Combining high-porosity 10 µm frits and 20 µm particles, the Styragel HMW columns minimize polymer shear effects. These state-of-the-art columns can be used at either ambient or elevated temperatures and exhibit excellent column lifetime.

### Calibration Curves for the Waters Styragel HMW Series of High Molecular Weight Columns

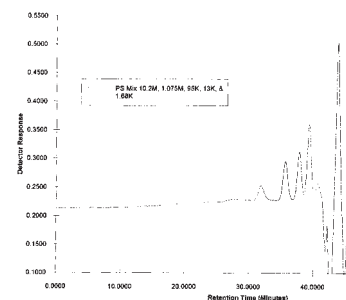


### Styragel HMW Columns are Optimized for Analysis of Shear-sensitive, Ultra-high Molecular Weight Polymers

Column Bank: 2 Styragel HMW 7 and 2 Styragel HMW 6E  
 Column Dimensions: 7.8 x 300 mm  
 Polystyrene Stds: 14.4M, 3.84M, 330K, 34.5K, 3.25K  
 Flow Rate: 1 mL/min  
 Temperature: 145 °C  
 Solvent: TCB



Column Bank: 2 Styragel HMW 7 and 2 Styragel HMW 6E  
 Column Dimensions: 7.8 x 300 mm  
 Polystyrene Stds: 10.2M, 1.075M, 95K, 13K, 1.68K  
 Flow Rate: 1 mL/min  
 Temperature: 145 °C  
 Solvent: TCB



### Styragel HMW Columns (7.8 mm i.d. x 300 mm)

Effective Molecular Column	Part No. Weight Range	Part No. THF	Part No. DMF	Part No. Toluene
Styragel HMW 2	100-10,000	WAT054488	WAT054494	WAT054490
Styragel HMW 7	500,000-1 x 10 <sup>8</sup>	WAT044201	WAT044202	WAT044200
Styragel HMW 6E	5,000-1 x 10 <sup>7</sup>	WAT044204	WAT044205	WAT044203
Styragel Guard Column 4.6 x 30 mm		WAT054405	WAT054415	WAT054410

### Styragel HMW Columns (4.6 mm i.d. x 300 mm)

The same high performance as our conventional Styragel HMW columns with the added advantage of reducing your solvent consumption by two-thirds.

Effective Molecular Column	Part No. Weight Range	Part No. THF	Part No. DMF	Part No. Toluene
Styragel HMW 7	500,000-1 x 10 <sup>8</sup>	WAT046805	WAT046810	WAT046800
Styragel HMW 6E	5,000-1 x 10 <sup>7</sup>	WAT046820	WAT046825	WAT046815

\*System dead volume must be minimized for maximum column performance.



## HSPgel Columns for High Speed GPC Analysis

Waters HSPgel™ Column offering for high-speed GPC analysis, provides for accurate and precise molecular weight determination, increased sample throughput, and greatly reduced solvent consumption and disposal. Waters offers four series of high speed GPC columns.

- HSPgel HR series is used for high resolution, room temperature GPC
- HSPgel RT series for routine room temperature GPC
- HSPgel HT series for high temperature GPC
- HSPgel AQ series for aqueous SEC

The column dimension for all columns is 6.0 x 150 mm.

The HSPgel HR series is designed for high resolution, room temperature, organic polymer GPC. These columns come packed in THF and can be converted once to toluene, methylene chloride, or chloroform.

The HSPgel RT series are designed for room temperature, routine work of organic polymer GPC. These come packed in THF and can be converted multiple times from THF to toluene, chloroform, methylene chloride, DMF, DMSO etc.

The HSPgel HT series are designed for room temperature to high temperature (180 °C) organic GPC. The columns come shipped in either THF or ODCB. The ODCB packed column should be used for direct conversion to TCB. These columns can withstand multiple solvent switches.

The HSPgel AQ series are designed for room temperature analysis of water soluble polymers.



Name	Solvent	Particle Size	MW Range	Part No.
<b>Ultra-High Resolution GPC</b>				
HSPgel HR 1.0	THF	3 µm	100-1,000	186001741
HSPgel HR 2.0	THF	3 µm	500-10,000	186001742
HSPgel HR 2.5	THF	3 µm	1,000-20,000	186001743
HSPgel HR 3.0	THF	3 µm	2,000-60,000	186001744
HSPgel HR 4.0	THF	3 µm	10,000-400,000	186001745
HSPgel HR MB-L	THF	3 µm	500-700,000	186001746
HSPgel HR MB-M	THF	3,5 µm	1,000-4,000,000	186001747
<b>Room-Temperature GPC</b>				
HSPgel RT 1.0	THF	3 µm	100-1,000	186001749
HSPgel RT 2.0	THF	3 µm	500-10,000	186001750
HSPgel RT 2.5	THF	3 µm	1,000-20,000	186001751
HSPgel RT 3.0	THF	3 µm	2,000-60,000	186001752
HSPgel RT 4.0	THF	3 µm	10,000-400,000	186001753
HSPgel RT 5.0	THF	3 µm	25,000-4,000,000	186001754
HSPgel RT 6.0	THF	5 µm	50,000-10,000,000	186001755
HSPgel RT MB-L	THF	3 µm	100-10,000	186001757
HSPgel RT MB-L/M	THF	3 µm	500-400,000	186001758
HSPgel RT MB-M	THF	3 µm	1,000-4,000,000	186001759
HSPgel RT MB-H	THF	3,5 µm	5,000->10,000,000	186001760
<b>Aqueous GPC</b>				
HSPgel AQ 2.5	Water	4 µm	500-2,000	186001785
HSPgel AQ 3.0	Water	4 µm	1,000-60,000	186001786
HSPgel AQ 4.0	Water	6 µm	10,000-400,000	186001787
HSPgel AQ 5.0	Water	7 µm	50,000-4,000,000	186001788
HSPgel AQ 6.0	Water	9 µm	100,000->10,000,000	186001789
HSPgel AQ MB-H	Water	9 µm	500-10,000,000	186001790

Name	Solvent	Particle Size	MW Range	Part No.
<b>High-Temperature GPC</b>				
HSPgel HT 1.0	THF	5 µm	100-1,000	186001761
HSPgel HT 2.0	THF	5 µm	500-10,000	186001762
HSPgel HT 2.5	THF	5 µm	1,000-20,000	186001763
HSPgel HT 3.0	THF	5 µm	2,000-60,000	186001764
HSPgel HT 4.0	THF	5 µm	10,000-400,000	186001765
HSPgel HT 5.0	THF	5 µm	25,000-4,000,000	186001766
HSPgel HT 6.0	THF	5 µm	50,000-10,000,000	186001767
HSPgel HT 7.0	THF	5 µm	100,000->15,000,000	186001768
HSPgel HT MB-L	THF	5 µm	100-1,000	186001769
HSPgel HT MB-L/M	THF	5 µm	500-400,000	186001770
HSPgel HT MB-M	THF	5 µm	1,000-4,000,000	186001771
HSPgel HT MB-H	THF	5 µm	5,000->10,000,000	186001772
HSPgel HT 1.0	ODCB	5 µm	100-1,000	186001773
HSPgel HT 2.0	ODCB	5 µm	500-10,000	186001774
HSPgel HT 2.5	ODCB	5 µm	1,000-20,000	186001775
HSPgel HT 3.0	ODCB	5 µm	2,000-60,000	186001776
HSPgel HT 4.0	ODCB	5 µm	10,000-400,000	186001777
HSPgel HT 5.0	ODCB	5 µm	25,000-4,000,000	186001778
HSPgel HT 6.0	ODCB	5 µm	50,000-10,000,000	186001779
HSPgel HT 7.0	ODCB	5 µm	100,000->15,000,000	186001780
HSPgel HT MB-L	ODCB	5 µm	100-1,000	186001781
HSPgel HT MB-L/M	ODCB	5 µm	500-400,000	186001782
HSPgel HT MB-M	ODCB	5 µm	1,000-4,000,000	186001783
HSPgel HT MB-H	ODCB	5 µm	5,000->10,000,000	186001784

All columns are 6.0 x 150 mm

\* MW ranges for HR and RT are based on polystyrene chain lengths, and based on polyethylene oxide chain lengths for the AQ series.

\*\* Exclusion limits for AQ series extrapolated from highest MW PEO standard, (~900,000).

HR — High Resolution      MB — Mixed Bed      L/M — Low/Medium MW Range  
 RT — Room Temperature      L — Low MW Range      H — High MW Range  
 AQ — Aqueous      M — Medium MW Range

## Shodex GPC Columns

High efficiency columns for GPC analysis. Featuring a very reproducible styrene divinylbenzoyne technology, Shodex® GPC columns have more than twenty years of history and have been used by customers all over the world.

### K-800 Series (8 x 300 mm)

These are ultra-high efficiency columns designed for high resolution performance. They are available in THF, DMF or chloroform.

### HFIP-800 Series (8 x 300 mm)

These columns have the same high efficiency as the K series columns but are available in HFIP.

### HFIP-800 Series (8 mm i.d. x 300 mm)

Type	Polystyrene Exclusion Limit	Part No.
HFIP-803	$7 \times 10^4$	WAT035605
HFIP-806M (linear)	$(4 \times 10^7)$	WAT035611
HFIP-LG precolumn (8 x 50 mm)		WAT035612

### Shodex GPC Columns

Type	Polystyrene Exclusion Limit	Part No.
KF-800 (THF)		
KF-801	$1.5 \times 10^3$	WAT030697
KF-802	$5 \times 10^3$	WAT030698
KF-802.5	$2 \times 10^4$	WAT030699
KF-803	$7 \times 10^4$	WAT034100
KF-804	$4 \times 10^5$	WAT034101
KF-805	$4 \times 10^6$	WAT034102
KF-807	$(2 \times 10^8)$	WAT034104
KF-806M (linear)	$(4 \times 10^7)$	WAT034105
KF-G pre-column (4.6 x 10 mm)		WAT034106
K-800 (Chloroform)		
K-802.5	$2 \times 10^4$	WAT034109
K-803	$7 \times 10^4$	WAT034110
K-804	$4 \times 10^5$	WAT034111
K-805	$4 \times 10^6$	WAT034112
K-G precolumn (4.6 x 10 mm)		WAT035524
KD-800 (DMF)		
KD-801	$2.5 \times 10^3$	WAT034116
KD-802	$5 \times 10^3$	WAT034117
KD-802.5	$2 \times 10^4$	WAT034118
KD-803	$7 \times 10^4$	WAT034119
KD-804	$4 \times 10^5$	WAT034120
KD-806	$(4 \times 10^7)$	WAT034122
KD-807	$(2 \times 10^8)$	WAT034123
KD-806M (linear)	$(4 \times 10^7)$	WAT034124
KD-G precolumn (4.6 x 10 mm)		WAT034125

## Envirogel High-Resolution GPC Cleanup Columns

The Envirogel™ high-efficiency GPC cleanup columns are specifically designed to remove low volatility, high molecular weight interferences, such as lipids and natural resins, from environmental samples as specified in EPA Method 3640A\*. In the past, the cleanup procedure for environmental samples was performed on low-efficiency GPC columns based on packing particle diameters of 37-75 µm (200 to 400 mesh) Bio-Beads S-X resins. The high-efficiency Envirogel GPC Cleanup columns increase the speed of this process while simultaneously reducing solvent consumption.

### Envirogel GPC Cleanup Columns Packed in Methylene Chloride

Column	Dimensions	Part No.
Envirogel GPC Cleanup	19 x 150 mm	WAT036555
Envirogel GPC Cleanup	19 x 300 mm	WAT036554
Envirogel GPC Guard	4.6 x 30 mm	186001913

\* EPA Method 3640A requires both columns.

### Envirogel GPC Cleanup Columns Packed in Cyclohexane/Ethyl Acetate

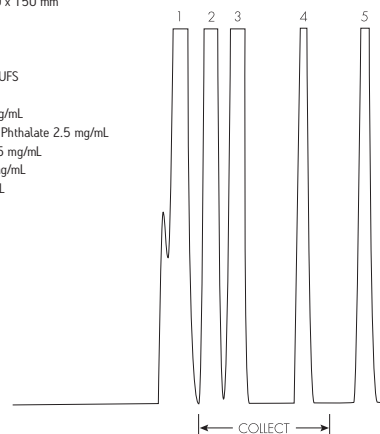
Column	Dimensions	Part No.
Envirogel GPC Cleanup	19 x 150 mm	186001915
Envirogel GPC Cleanup	19 x 300 mm	186001916
Envirogel GPC Guard	4.6 x 30 mm	186001914

\* EPA Method 3640A requires both columns.

### Column Optimization

Column: Two Envirogel GPC columns  
19 x 300 mm and 19 x 150 mm  
Sample: 2000 µL  
Solvent: Methylene Chloride  
Flow Rate: 5 mL/min  
Detection: UV at 254 nm, 1.5 AUFS

1. Corn oil, 62.5 mg/mL
2. Bis(2-Ethylhexyl) Phthalate 2.5 mg/mL
3. Methoxychlor, 0.5 mg/mL
4. Perylene, 0.05 mg/mL
5. Sulfur, 0.2 mg/mL



For optimum capacity and resolution, a 150 mm column is used in series with the 300 mm column. The use of both the 150 mm column and the 300 mm column provides maximum loading capacity while the 300 mm column provides maximum throughput and reduction in solvent consumption when used alone.

## Preparative GPC Columns

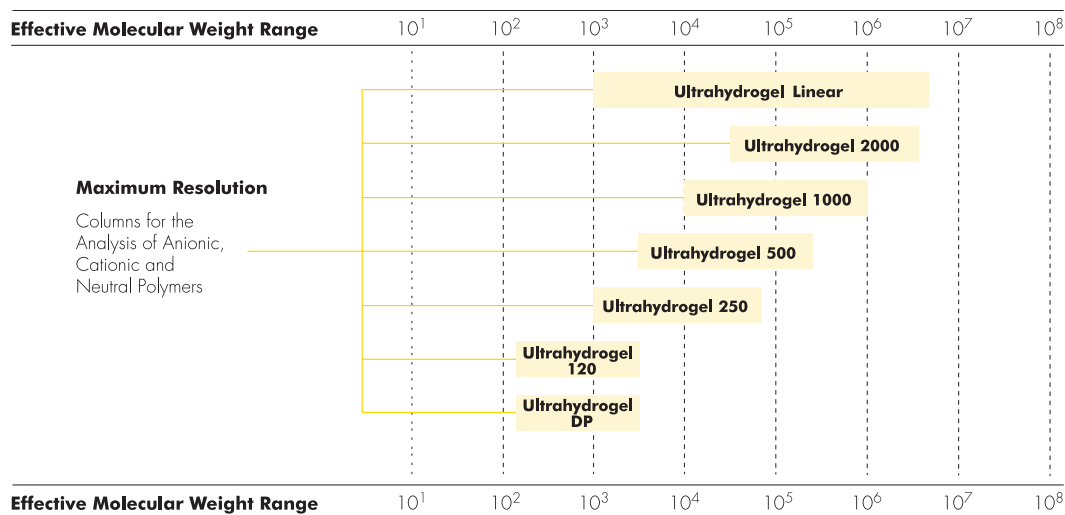
### Ultrastyrigel Columns (19 mm i.d. x 300 mm)

For high resolution preparative applications, these columns are available in toluene or THF.

Pore Size	Effective Molecular Weight Range	Flow Rate mL/Min	Part No.	Part No.
			Toluene	THF
100Å	50-1,500	4-10	WAT025866	WAT025859
500Å	100-10,000	4-10	WAT025867	WAT025860
103Å	200-30,000	4-10	WAT025868	WAT025861
104Å	5,000-600,000	4-10	WAT025869	WAT025862
105Å	50,000-4M	4-10	WAT025870	WAT025863
106Å	200,000-10M	4-10	WAT025871	WAT025864
Linear	2,000-4M	4-10	WAT025872	WAT025865

[Effective linear functional range: 2K-4M]

## Aqueous SEC Column Selection Guide



## Eluent Selection—Aqueous SEC with Ultrahydrogel Columns

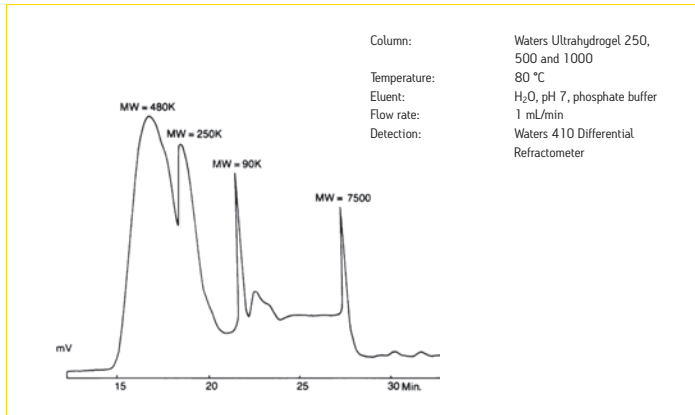
Class	Polymer	Eluent
Nonionic	Polyethylene Oxide Polyethylene Glycol Polysaccharides Pullulans Dextrans Cellulosics (H <sub>2</sub> O Soluble) Polyvinyl Alcohol Polyacrylamide	0.10M NaNO <sub>3</sub>
Nonionic Hydrophobic	Polyvinyl Pyrrolidone	80/20, 0.10M NaNO <sub>3</sub> /CH <sub>3</sub> CN
Anionic	Polyacrylic Acid (Na) Polyalginic Acid (Na) Hyaluronic Acid Carrageenan	0.10M NaNO <sub>3</sub>
Anionic Hydrophobic	Polystyrene Sulfonate (Na) Lignin, Sulfonated	80/20, 0.10M NaNO <sub>3</sub> /CH <sub>3</sub> CN
Cationic	DEAE Dextran Polyvinylamine Polyepiamine N-Acetylglucosamine	0.80M NaNO <sub>3</sub> 0.10M TEA 0.10M TEA/1% HOAc
Cationic Hydrophobic	Polyethyleneimine Poly (n-Methyl-2-Vinyl Pyridinium) Lysozyme Chitosan Polylysine Peptides	0.50M NaOAc/0.50M HOAc 0.5M CH <sub>3</sub> COOH/0.3M Na <sub>2</sub> SO <sub>4</sub> 0.50M HOAc/0.30M Na <sub>2</sub> SO <sub>4</sub> 5% NaH <sub>2</sub> PO <sub>4</sub> with 3% CH <sub>3</sub> CN(pH 4.0) 0.10% TFA/40% CH <sub>3</sub> CN
Amphoteric	Collagen/Gelatin	80/20, 0.10M NaNO <sub>3</sub> /CH <sub>3</sub> CN

## Ultrahydrogel Columns

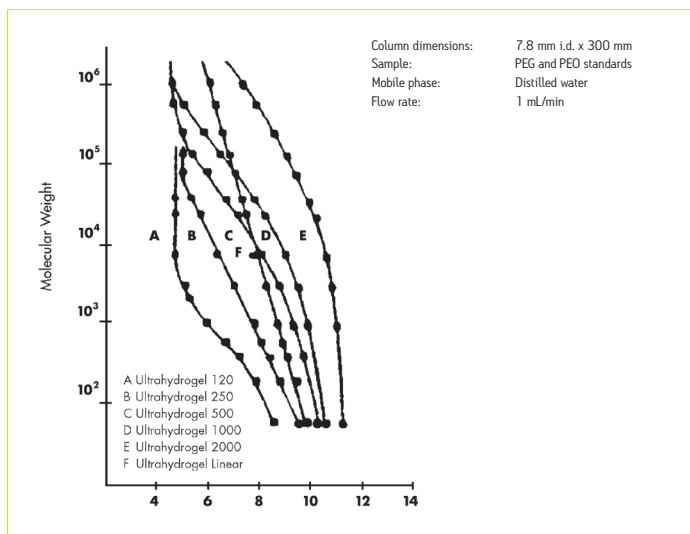
Packed with hydroxylated polymethacrylate-based gel, Waters Ultrahydrogel™ SEC columns are ideal for the analysis of aqueous-soluble samples, such as oligomers; oligosaccharides; polysaccharides; and cationic, anionic, and amphoteric polymers. Measuring 7.8 x 300 mm, these high-resolution columns offer many advantages over conventional aqueous SEC columns, such as:

- A wide pH range (2-12)
- Compatibility with high concentrations of organic solvents (up to 20% organic, 50% organic if the mobile phase is introduced by gradient)
- Greater flexibility for the mobile phase
- Minimal non-size-exclusion effects

### Gelatin Sample



### Ultrahydrogel Columns Calibration Curves



### Ultrahydrogel Columns (7.8 x 300 mm)

Column	Pore Size	Exclusion Limit	Part No.
Ultrahydrogel 120	120 Å	5 x 10 <sup>3</sup>	WAT011520
Ultrahydrogel 250	250 Å	8 x 10 <sup>4</sup>	WAT011525
Ultrahydrogel 500	500 Å	4 x 10 <sup>5</sup>	WAT011530
Ultrahydrogel 1000	1000 Å	1 x 10 <sup>6</sup>	WAT011535
Ultrahydrogel 2000	2000 Å	7 x 10 <sup>6</sup>	WAT011540
Ultrahydrogel Linear	Blend	7 x 10 <sup>6</sup>	WAT011545
Ultrahydrogel DP*	120 Å	5 x 10 <sup>3</sup>	WAT011550
Ultrahydrogel Guard Column	N/A	N/A	WAT011565
Ultrahydrogel Guard Column DP*	N/A	N/A	WAT011570

\* DP = Degree of Polymerization, choice of column when working with glucose oligomers.

# Calibration Standards

## Organic Calibration Standards

### Organic Standards Kits

Description	Qty./ MW	Part No.
Polystyrene Kit Low-Mid MW*	4 x 10 <sup>2</sup> /10 g, 5.3 x 10 <sup>2</sup> /10 g, 9.5 x 10 <sup>2</sup> /10 g, 2.8 x 10 <sup>3</sup> /5 g, 6.4 x 10 <sup>3</sup> /5 g, 1 x 10 <sup>4</sup> /5 g, 1.7 x 10 <sup>4</sup> /5 g, 4.3 x 10 <sup>4</sup> /5 g, 1.1 x 10 <sup>5</sup> /5 g, 1.8 x 10 <sup>5</sup> /5 g	WAT011588
Polystyrene Kit Mid-High MW	4.3 x 10 <sup>5</sup> /5 g, 7.8 x 10 <sup>5</sup> /5 g, 1.3 x 10 <sup>6</sup> /1 g, 2.8 x 10 <sup>6</sup> /1 g, 3.6 x 10 <sup>6</sup> /1 g, 4.3 x 10 <sup>6</sup> /1 g, 5.2 x 10 <sup>6</sup> /1 g, 6.2 x 10 <sup>6</sup> /1 g, 8.4 x 10 <sup>6</sup> /1 g, 2 x 10 <sup>7</sup> /1 g	WAT011610
Polystyrene Kit SL-105	5.8 x 10 <sup>2</sup> , 9.5 x 10 <sup>2</sup> , 1.2 x 10 <sup>3</sup> , 1.8 x 10 <sup>3</sup> , 2.47 x 10 <sup>3</sup> , 3.77 x 10 <sup>3</sup> , 5.1 x 10 <sup>3</sup> , 7.6 x 10 <sup>3</sup> , 1.25 x 10 <sup>4</sup> , 1.7 x 10 <sup>4</sup>	500 mg/each WAT034208
Polystyrene Kit SM-105	1.2 x 10 <sup>3</sup> , 3.25 x 10 <sup>3</sup> , 1.02 x 10 <sup>4</sup> , 2.8 x 10 <sup>4</sup> , 6.8 x 10 <sup>4</sup> , 1.95 x 10 <sup>5</sup> , 4.9 x 10 <sup>5</sup> , 1.08 x 10 <sup>6</sup> , 1.75 x 10 <sup>6</sup> , 2.75 x 10 <sup>6</sup>	500 mg/each WAT034209
Polystyrene Kit SH-75	4.5 x 10 <sup>5</sup> , 1.27 x 10 <sup>6</sup> , 2.3 x 10 <sup>6</sup> , 3.26 x 10 <sup>6</sup> , 4.34 x 10 <sup>6</sup> , 8 x 10 <sup>6</sup> , 1.5 x 10 <sup>7</sup>	500 mg/each WAT034210
Polymethylmethacrylate Mid MW Kit	2.4 x 10 <sup>3</sup> , 9.5 x 10 <sup>3</sup> , 3.1 x 10 <sup>4</sup> , 5.2 x 10 <sup>4</sup> , 1 x 10 <sup>5</sup> , 1.7 x 10 <sup>5</sup> , 2.7 x 10 <sup>5</sup> , 4.91 x 10 <sup>5</sup> , 7.3 x 10 <sup>5</sup> , 1 x 10 <sup>6</sup>	500 mg/each WAT035706
Polymethylmethacrylate Low MW Kit	1 x 10 <sup>3</sup> , 1.7 x 10 <sup>3</sup> , 2.5 x 10 <sup>3</sup> , 3.5 x 10 <sup>3</sup> , 5 x 10 <sup>3</sup> , 7 x 10 <sup>3</sup> , 1 x 10 <sup>4</sup> , 1.3 x 10 <sup>4</sup> , 2 x 10 <sup>4</sup> , 3 x 10 <sup>4</sup>	500 mg/each WAT035707
Polybutadiene Kit	1 x 10 <sup>3</sup> , 3 x 10 <sup>3</sup> , 7 x 10 <sup>3</sup> , 1 x 10 <sup>4</sup> , 3 x 10 <sup>4</sup> , 7 x 10 <sup>4</sup> , 1 x 10 <sup>5</sup> , 3 x 10 <sup>5</sup> , 7 x 10 <sup>5</sup> , 1.1 x 10 <sup>6</sup>	500 mg/each WAT035709
Polyisoprene Kit	1 x 10 <sup>3</sup> , 3 x 10 <sup>3</sup> , 1 x 10 <sup>4</sup> , 3 x 10 <sup>4</sup> , 7 x 10 <sup>4</sup> , 1 x 10 <sup>5</sup> , 3 x 10 <sup>5</sup> , 5 x 10 <sup>5</sup> , 1 x 10 <sup>6</sup> , 3 x 10 <sup>6</sup>	500 mg/each WAT035708

\* Approximate molecular weights

### Polystyrene (PS) Organic Standards (Individual Standard)

Approximate Molecular Weight Range		Qty.	Part No.
LS*	GPC		
—	4 x 10 <sup>2</sup>	10 g	WAT011590
—	5.3 x 10 <sup>2</sup>	10 g	WAT011592
—	9.5 x 10 <sup>2</sup>	10 g	WAT011594
2.8 x 10 <sup>3</sup>	2.8 x 10 <sup>3</sup>	5 g	WAT011596
6.2 x 10 <sup>3</sup>	6.4 x 10 <sup>3</sup>	5 g	WAT011598
1.03 x 10 <sup>4</sup>	1.01 x 10 <sup>4</sup>	5 g	WAT011600
1.67 x 10 <sup>4</sup>	1.73 x 10 <sup>4</sup>	5 g	WAT011602
4.39 x 10 <sup>4</sup>	4.30 x 10 <sup>4</sup>	5 g	WAT011604
1.07 x 10 <sup>5</sup>	1.06 x 10 <sup>5</sup>	5 g	WAT011606
1.86 x 10 <sup>5</sup>	1.84 x 10 <sup>5</sup>	5 g	WAT011608
4.22 x 10 <sup>5</sup>	4.27 x 10 <sup>5</sup>	5 g	WAT011612
7.75 x 10 <sup>5</sup>	7.91 x 10 <sup>5</sup>	5 g	WAT011614
1.26 x 10 <sup>6</sup>	1.30 x 10 <sup>6</sup>	1 g	WAT011616
2.86 x 10 <sup>6</sup>	2.80 x 10 <sup>6</sup>	1 g	WAT011618
3.84 x 10 <sup>6</sup>	3.61 x 10 <sup>6</sup>	1 g	WAT011620
4.48 x 10 <sup>6</sup>	4.27 x 10 <sup>6</sup>	1 g	WAT011622
5.48 x 10 <sup>6</sup>	5.20 x 10 <sup>6</sup>	1 g	WAT011624
6.77 x 10 <sup>6</sup>	6.20 x 10 <sup>6</sup>	1 g	WAT011626
8.42 x 10 <sup>6</sup>	—	1 g	WAT011628
2.0 x 10 <sup>7</sup>	—	1 g	WAT011630

\* Light scattering

### ReadyCal Polystyrene Standards

Thirty autosampler vials which contain 4 polystyrene standards per vial. There are three separate molecular weight ranges in each kit, 10 units of each of the three molecular weight standards. Just add solvent to the vial, let stand for two hours, shake gently and load into your autosampler for analysis. Each kit comes with detailed instructions for proper usage.

Type	Approximate Standards	Molecular Weight	Part No.
ReadyCal, 4 mL Autosampler Vial	12	4 x 10 <sup>2</sup> x 2 x 10 <sup>6</sup>	WAT058930
ReadyCal, 2 mL Autosampler Vial	12	4 x 10 <sup>2</sup> x 2 x 10 <sup>6</sup>	WAT058931

## Aqueous Calibration Standards

### Aqueous Standards Kits

Description	Qty./ MW	Part No.
Pullulan Kit	* 5.8 x 10 <sup>3</sup> , 1.22 x 10 <sup>4</sup> , 2.37 x 10 <sup>4</sup> , 1 x 10 <sup>5</sup> , 1.86 x 10 <sup>5</sup> , 3.8 x 10 <sup>5</sup> , 8.53 x 10 <sup>5</sup>	200 mg/each WAT034207
Dextran Kit	5 x 10 <sup>3</sup> , 1.2 x 10 <sup>4</sup> , 2.4 x 10 <sup>4</sup> , 4.8 x 10 <sup>4</sup> , 1.48 x 10 <sup>5</sup> , 2.73 x 10 <sup>5</sup> , 4.1 x 10 <sup>5</sup> , 7.5 x 10 <sup>5</sup>	500 mg/each WAT054392
Polyethyleneoxide (PEO) Kit	2.4 x 4 x 10 <sup>4</sup> , 8 x 10 <sup>4</sup> , 1.6 x 10 <sup>5</sup> , 3.4 x 10 <sup>5</sup> , 5.7 x 10 <sup>5</sup> , 8.5 x 10 <sup>5</sup>	500 mg/each WAT011572
Polyethylene Glycol (PEG) Kit	1 x 10 <sup>2</sup> , 2 x 10 <sup>2</sup> , 4 x 10 <sup>2</sup> , 6 x 10 <sup>2</sup> , 1 x 10 <sup>3</sup> , 1.5 x 10 <sup>3</sup> , 4.3 x 10 <sup>3</sup> , 7 x 10 <sup>3</sup> , 1.3 x 10 <sup>4</sup> , 2.2 x 10 <sup>4</sup>	1 gram/each WAT035711
Polyacrylic Acid Kit	1 x 10 <sup>3</sup> , 3 x 10 <sup>3</sup> , 7 x 10 <sup>3</sup> , 1.5 x 10 <sup>4</sup> , 3 x 10 <sup>4</sup> , 7 x 10 <sup>4</sup> , 1 x 10 <sup>5</sup> , 3 x 10 <sup>5</sup> , 7 x 10 <sup>5</sup> , 1 x 10 <sup>6</sup>	250 mg/each WAT035714

\* Approximate molecular weights

### Polyethyleneoxide (PEO) Aqueous Standards

Approximate Molecular Weight Range		Qty.	Part No.
LS*	GPC		
2.5 x 10 <sup>4</sup>	2.4 x 10 <sup>4</sup>	0.5 g	WAT011574
4.0 x 10 <sup>4</sup>	4.0 x 10 <sup>4</sup>	0.5 g	WAT011576
7.3 x 10 <sup>4</sup>	7.9 x 10 <sup>4</sup>	0.5 g	WAT011578
1.5 x 10 <sup>5</sup>	1.6 x 10 <sup>5</sup>	0.5 g	WAT011580
2.8 x 10 <sup>5</sup>	3.4 x 10 <sup>5</sup>	0.5 g	WAT011582
6.6 x 10 <sup>5</sup>	5.7 x 10 <sup>5</sup>	0.5 g	WAT011584
8.5 x 10 <sup>5</sup>	8.5 x 10 <sup>5</sup>	0.5 g	WAT011586

\* Light scattering