

# PROCESS DEVELOPMENT PRODUCTS AND BULK RESINS FOR LABORATORY SCALE PURIFICATION

PROCESS DEVELOPMENT & RESINS

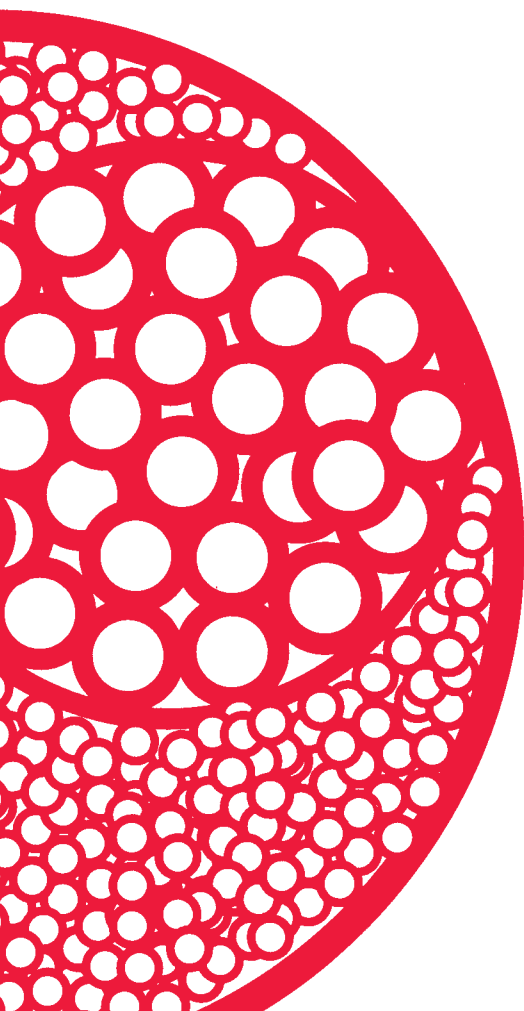
- ToyoScreen PROCESS DEVELOPMENT COLUMNS
- RoboColumn PROCESS DEVELOPMENT COLUMNS
- MiniChrom PROCESS DEVELOPMENT COLUMNS
- TOYOPEARL AND TSKgel LabPAK
- TOYOPEARL AND TSKgel BULK RESINS

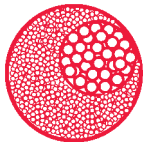
## ≡ TOSOH FACT

Tosoh Bioscience offers a range of technical support services to our TSKgel, ToyoScreen, and TOYOPEARL chromatography products.

Whether you need help developing an HPLC assay for the analysis of a new therapeutic target, want to know how to monitor drug metabolites in the human body or need regulatory files to support a submission to the FDA, our technical support specialists will provide assistance in all of these areas and more.

We offer on-site training and application-specific seminars and are committed to providing prompt and courteous service for these and other requests.





## ToyoScreen PROCESS DEVELOPMENT COLUMNS

ToyoScreen Process Development columns are easy-to-use, pre-packed columns containing the most popular TOYOPEARL resins. These columns provide a convenient, low-cost method for the evaluation of TOYOPEARL ligand chemistries. ToyoScreen Process Development columns are available in volumes of 1 mL and 5 mL for affinity, ion exchange, mixed-mode and hydrophobic interaction chromatography. See the chapter on bulk resins for detailed information on TOYOPEARL resins.

### SCREENING

Historically, resin screening was accomplished by manually packing various bulk resins into small columns requiring a significant investment in time and cost. In order to improve the efficiency of resin screening experiments, pre-packed ToyoScreen Process Development columns were developed for the evaluation of different TOYOPEARL resins.

### SCALABILITY

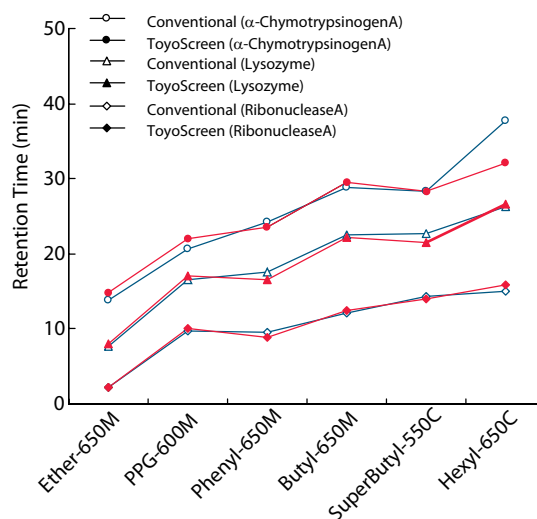
Initial results from resin screening and optimization with ToyoScreen columns can accurately predict the separation behavior at larger scales. **FIGURE 1** illustrates the similar retention time behavior between 1 mL ToyoScreen columns and conventional 7.5 mm ID x 7.5 cm L analytical columns. Additionally, **FIGURE 2** depicts a practical antibody scale up in which conditions were set using a 1 mL ToyoScreen column and applied to a 10 mL semi-preparative column with a different inner diameter and length. Similar resolution results are predicted by the following equation:

$$Rs \propto \frac{1}{dp} \frac{z^{1/2}}{u^{1/2} (g(V_t - V_o))^{1/2}}$$

### FEATURES

- Pre-packed columns
- 1 mL and 5 mL bed volume
- Cartridge design
- Ready to connect to ÄKTA, FPLC and HPLC systems
- Six pieces offered in mixed or single chemistry

**FIGURE 1**  
Comparison of selectivity between ToyoScreen and conventional column



Columns: ToyoScreen (6.4 mm ID x 3 cm L), Conventional Column (7.5 mm ID x 7.5 cm L);

Eluent A: 0.1 mol/L phosphate buffer + 1.8 mol/L sodium sulfate (pH 7.0),

Eluent B: 0.1 mol/L phosphate buffer (pH 7.0); Flow rate: 1 mL/min

Gradient: 30 min linear; Inj. Vol.: 50 µL; Samples: Ribonuclease A, Lysozyme, α-Chymotrypsinogen, 1 mg/mL

Retention time of conventional column was plotted after converting following equation: plotted value = actual measurement value - 4.82

### METHOD OPTIMIZATION

Besides the determination of what sticks during resin screening experiments, ToyoScreen Process Development columns can be used to quickly establish optimum elution conditions. Varying pH, salt type, salt gradients and flow rate are common experimental parameters explored. The effect of varying salt type and pH are shown in **FIGURES 3 & 4** for anti-TSH in cell culture supernatant on ToyoScreen Phenyl-650M.

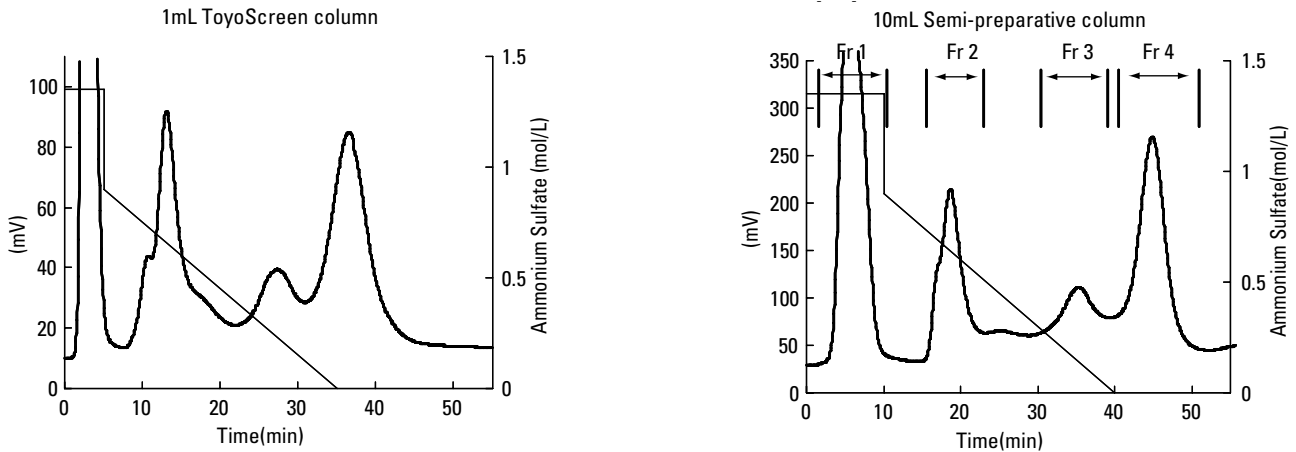
### BENEFITS

- Easy to set up and screen an entire resin series for a specific chromatographic mode
- For sample limited applications with up to milligram purifications
- Provides low cost, efficient alternative to hand packing with bulk resin
- Seamless integration into any platform
- For cost savings in screening or process experiments

# PROCESS DEVELOPMENT

## APPLICATIONS - ToyoScreen PROCESS DEVELOPMENT COLUMNS

**FIGURE 2**  
Comparison chromatograms between ToyoScreen and semi-preparative columns

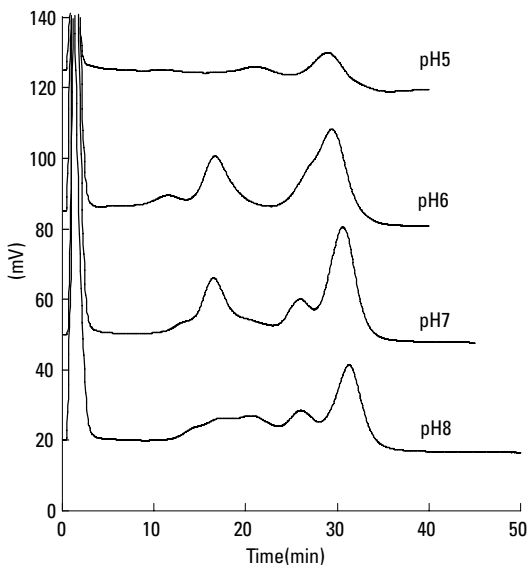


Packing: TOYOPEARL Phenyl-650M; Eluent: (A) 0.1 mol/L phosphate buffer containing 1.8 mol/L  $(NH_4)_2SO_4$ , pH 7.0 (B) 0.1 mol/L phosphate buffer, pH7.0; Sample: Anti-TSH from cell culture supernatant (x4 diluted)

	1 mL ToyoScreen	10 mL Semi-preparative
Column Dimensions:	6.4 mm ID x 3 cm L	14.6 mm ID x 6 cm L
Injection Volume:	500 $\mu$ L	5000 $\mu$ L
Flow rate:	0.5 mL/min; 0.5 CV/min; 93 cm/h	2.5 mL/min; 0.25 CV/min; 90 cm/h
Gradient Profile:	25% B; 0-5 min (isocratic) 50% B: 5 min (step) 50% to 100% B; 5-35 min (linear)	25% B; 0-10 min (isocratic) 50% B: 10 min (step) 50% to 100% B; 10-40 min (linear)
Gradient Slope*:	0.06 M/mL	0.012 M/mL

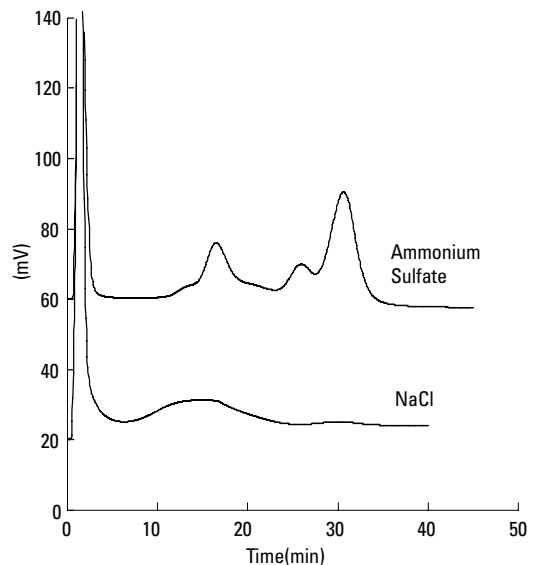
\* The gradient slope is the change in ionic strength per unit volume. Gradient volume is the product of flow rate and gradient time.

**FIGURE 3**  
Optimizing eluent pH in HIC



Column: ToyoScreen Phenyl-650M (1 mL); Eluent A: 0.1 mol/L phosphate buffer + 1.8 mol/L ammonium sulfate (pH7.0); Eluent B: 0.1 mol/L phosphate buffer (pH 7.0); Flow rate: 1 mL/min; Gradient: 30 min linear (30 CV); Inj. Vol.: 200  $\mu$ L; Sample: Cell culture supernatant (x4 diluted) (antibody: Anti-TSH)

**FIGURE 4**  
Optimizing salt conditions in HIC



Column: ToyoScreen Phenyl-650M (1 mL); Eluent A: 0.1 mol/L phosphate buffer containing 1.8 mol/L each salt (pH7.0); Eluent B: 0.1 mol/L phosphate buffer (pH 7.0); Flow rate: 1 mL/min; Gradient: 30 min linear (30 CV); Inj. Vol.: 200  $\mu$ L; Sample: Cell culture supernatant (x 4 diluted) (antibody: Anti-TSH)


**ORDERING INFORMATION**

<i>Part #</i>	<i>Description</i>	<i>Package description</i>	<i>Part #</i>	<i>Description</i>	<i>Package description</i>
<b>Ion Exchange</b>			<b>Hydrophobic Interaction</b>		
0021360	ToyoScreen DEAE-650M, 1 mL	1 mL x 6 ea	0021372	ToyoScreen Ether-650M, 1 mL	1 mL x 6 ea
0021361	ToyoScreen DEAE-650M, 5 mL	5 mL x 6 ea	0021373	ToyoScreen Ether-650M, 5 mL	5 mL x 6 ea
0021362	ToyoScreen SuperQ-650M, 1 mL	1 mL x 6 ea	0021892	ToyoScreen Phenyl-600M, 1 mL	1 mL x 6 ea
0021363	ToyoScreen SuperQ-650M, 5 mL	5 mL x 6 ea	0021893	ToyoScreen Phenyl-600M, 5 mL	5 mL x 6 ea
0021364	ToyoScreen QAE-550C, 1 mL	1 mL x 6 ea	0021374	ToyoScreen Phenyl-650M, 1 mL	1 mL x 6 ea
0021365	ToyoScreen QAE-550C, 5 mL	5 mL x 6 ea	0021375	ToyoScreen Phenyl-650M, 5 mL	5 mL x 6 ea
0021992	ToyoScreen Q-600C AR, 1 mL	1 mL x 6 ea	0021494	ToyoScreen Butyl-600M, 1 mL	1 mL x 6 ea
0021993	ToyoScreen Q-600C AR, 5 mL	5 mL x 6 ea	0021495	ToyoScreen Butyl-600M, 5 mL	5 mL x 6 ea
0021859	ToyoScreen GigaCap Q-650M, 1 mL	1 mL x 6 ea	0021376	ToyoScreen Butyl-650M, 1 mL	1 mL x 6 ea
0021860	ToyoScreen GigaCap Q-650M, 5 mL	5 mL x 6 ea	0021377	ToyoScreen Butyl-650M, 5 mL	5 mL x 6 ea
0022873	ToyoScreen GigaCap DEAE-650M, 1 mL	1 mL x 6 ea	0021378	ToyoScreen Hexyl-650C, 1 mL	1 mL x 6 ea
0022872	ToyoScreen GigaCap DEAE-650M, 5 mL	5 mL x 6 ea	0021379	ToyoScreen Hexyl-650C, 5 mL	5 mL x 6 ea
0023443	ToyoScreen NH <sub>2</sub> -750F	1 mL x 6 ea	0021380	ToyoScreen PPG-600M, 1 mL	1 mL x 6 ea
0023444	ToyoScreen NH <sub>2</sub> -750F	5 mL x 6 ea	0021381	ToyoScreen PPG-600M, 5 mL	5 mL x 6 ea
0021870	ToyoScreen MegaCapII SP-550EC, 1 mL	1 mL x 6 ea	0021382	ToyoScreen SuperButyl-550C, 1 mL	1 mL x 6 ea
0021871	ToyoScreen MegaCapII SP-550EC, 5 mL	5 mL x 6 ea	0021383	ToyoScreen SuperButyl-550C, 5 mL	5 mL x 6 ea
0021366	ToyoScreen CM-650M, 1mL	1 mL x 6 ea	0021398	ToyoScreen HIC Mix Pack, 1 mL	1 mL x 6 Grades x 1 ea (PPG-600M, Butyl-600M/-650M, Phenyl-600M/-650M, Hexyl-650C)
0021367	ToyoScreen CM-650M, 5mL	5 mL x 6 ea	0021399	ToyoScreen HIC Mix Pack, 5 mL	5 mL x 6 Grades x 1 ea (PPG-600M, Butyl-600M/-650M, Phenyl-600M/-650M, Hexyl-650C)
0021951	ToyoScreen GigaCap CM-650M, 1 mL	1 mL x 6 ea	<b>Affinity</b>		
0021952	ToyoScreen GigaCap CM-650M, 5 mL	5 mL x 6 ea	0023430	ToyoScreen AF-rProtein A HC-650F, 1 mL	1 mL x 5 ea
0021368	ToyoScreen SP-650M, 1mL	1 mL x 6 ea	0023431	ToyoScreen AF-rProtein A HC-650F, 5 mL	5 mL x 1 ea
0021369	ToyoScreen SP-650M, 5mL	5 mL x 6 ea	0023432	ToyoScreen AF-rProtein A HC-650F, 5 mL	5 mL x 5 ea
0021370	ToyoScreen SP-550C, 1mL	1 mL x 6 ea	0022809	ToyoScreen AF-rProtein A-650F, 1 mL	1 mL x 5 ea
0021371	ToyoScreen SP-550C, 5mL	5 mL x 6 ea	0022810	ToyoScreen AF-rProtein A-650F, 5 mL	5 mL x 1 ea
0021868	ToyoScreen GigaCap S-650M, 1 mL	1 mL x 6 ea	0022811	ToyoScreen AF-rProtein A-650F, 5 mL	5 mL x 5 ea
0021869	ToyoScreen GigaCap S-650M, 5 mL	5 mL x 6 ea	0021386	ToyoScreen AF-Blue HC-650M, 1 mL	1 mL x 6 ea
0021392	ToyoScreen IEC Anion Mix Pack, 1 mL	1 mL x 5 Grades (DEAE-650M, SuperQ-650M, QAE-550C, GigaCap Q-650M, Q-600C AR)	0021387	ToyoScreen AF-Blue HC-650M, 5 mL	5 mL x 6 ea
0021393	ToyoScreen IEC Anion Mix Pack, 5 mL	5 mL x 5 Grades (DEAE-650M, SuperQ-650M, QAE-550C, GigaCap Q-650M, Q-600C AR)	0021384	ToyoScreen AF-Chelate-650M, 1 mL	1 mL x 6 ea
0021394	ToyoScreen IEC Cation Mix Pack, 1 mL	1 mL x 5 Grades (CM-650M, SP-650M, SP-550C, GigaCap CM-650M /S-650M)	0021385	ToyoScreen AF-Chelate-650M, 5 mL	5 mL x 6 ea
0021395	ToyoScreen IEC Cation Mix Pack, 5 mL	5 mL x 5 Grades (CM-650M, SP-650M, SP-550C, GigaCap CM-650M /S-650M)	0021390	ToyoScreen AF-Heparin HC-650M, 1 mL	1 mL x 6 ea
0021396	ToyoScreen IEC Mix Pack, 1 mL	1 mL x 6 Grades x 1 ea (GigaCap Q-650M/ CM-650M/S-650M, SuperQ-650M, Q-600C AR)	0021391	ToyoScreen AF-Heparin HC-650M, 5 mL	5 mL x 6 ea
0021397	ToyoScreen IEC Mix Pack, 5 mL	5 mL x 6 Grades x 1 ea (GigaCap Q-650M/ CM-650M/S-650M, SuperQ-650M, Q-600C AR)	0021388	ToyoScreen AF-Red-650M, 1 mL	1 mL x 6 ea
			0021389	ToyoScreen AF-Red-650M, 5 mL	5 mL x 6 ea
<b>Mixed-Mode</b>			<b>ToyoScreen accessories</b>		
0022824	ToyoScreen MX-Trp-650M, 1 mL	1 mL x 6 ea	0021400	ToyoScreen column holder	
0022825	ToyoScreen MX-Trp-650M, 5 mL	5 mL x 6 ea			

# PROCESS DEVELOPMENT

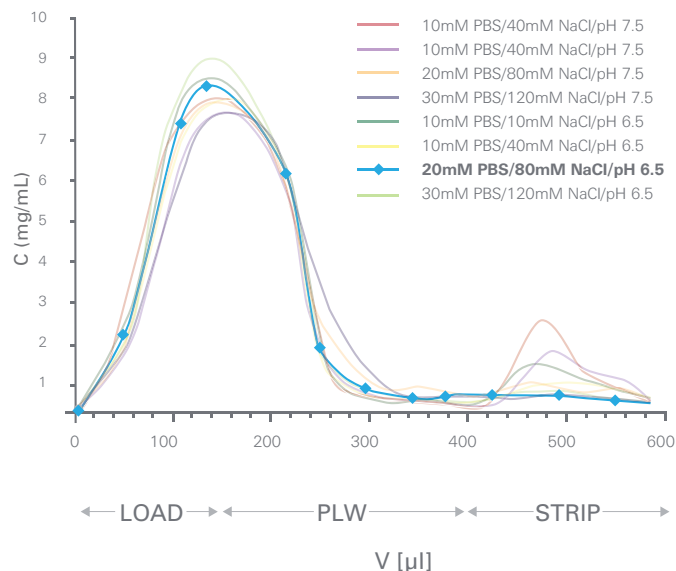
## RoboColumns for TOYOPEARL

Tosoh Bioscience offers TOYOPEARL media now also in the well-known RoboColumn® format packed by Atoll GmbH. RoboColumns are miniaturized chromatographic columns pre-packed with the most popular TOYOPEARL ion exchange, mixed-mode, hydrophobic interaction or affinity media. The columns are available in different volumes and can be operated with a robotic liquid handling system. This approach allows automated high-throughput, small-scale chromatographic separations of protein samples by running up to eight individual columns simultaneously.

RoboColumns are available in two formats with 200 µL (bed height of 10 mm) and 600 µL (bed height of 30 mm) resin volume, respectively. They are supplied in a row of eight units pre-packed with the same TOYOPEARL resin and sealed with two removable silicon cover seals for proper storage. A 96-well array plate is available to arrange the up to 96 RoboColumn units.

Figure 1 shows a screening experiment to optimize the parameters for the intermediate flow-through anion exchange step in a mAb purification platform. Protein binding of a Protein A capture eluate on RoboColumns packed with TOYOPEARL SuperQ-650M was analyzed by varying salt concentration and pH of loading and washing buffer. Best results were achieved using 20 mmol/L sodium phosphate, 80 mmol/L sodium chloride, pH 6.5.

**FIGURE 5**  
Optimization of anion exchange conditions



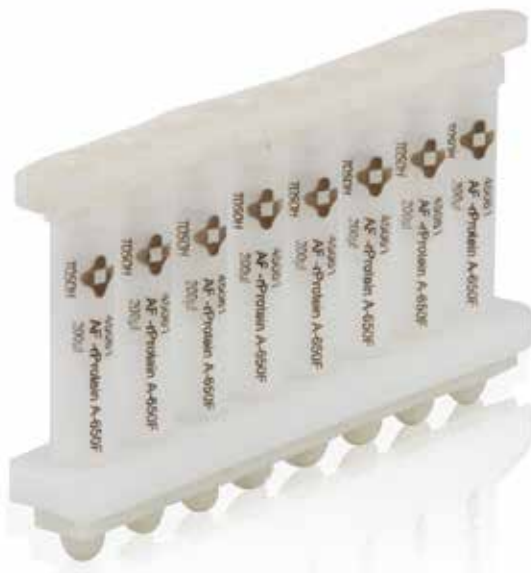
Elution profile of a protein A capture eluate on RoboColumns packed with Toyopearl SuperQ-650M at various conditions. Data kindly provided by T. Schröder, Atoll GmbH.

## FEATURES

- Pre-packed columns for use with robotic systems
- 200 and 600 µl bed volume
- Miniaturized column format

## BENEFITS

- High throughput parallel chromatography
- Fast resin screening and evaluation of design space
- Low consumption of sample and buffers



**ORDERING INFORMATION**

Part #	Description	Package description
<b>ToyoScreen RoboColumns for fast automated screening of resins</b>		
0045099		Array Plate

**Gel Filtration / Desalting**

0045071	RoboColumn HW-40F	0.2 mL*8 cols
0045072	RoboColumn HW-40F	0.6 mL*8 cols

**Ion Exchange**

0045023	RoboColumn GigaCap S-650S	0.2 mL*8 cols
0045024	RoboColumn GigaCap S-650S	0.6 mL*8 cols
0045001	RoboColumn GigaCap S-650M	0.2 mL*8 cols
0045002	RoboColumn GigaCap S-650M	0.6 mL*8 cols
0045025	RoboColumn GigaCap Q-650S	0.2 mL*8 cols
0045026	RoboColumn GigaCap Q-650S	0.6 mL*8 cols
0045002	RoboColumn GigaCap S-650M	0.6 mL*8 cols
0045003	RoboColumn GigaCap Q-650M	0.2 mL*8 cols
0045004	RoboColumn GigaCap Q-650M	0.6 mL*8 cols
0045005	RoboColumn GigaCap CM-650M	0.2 mL*8 cols
0045006	RoboColumn GigaCap CM-650M	0.6 mL*8 cols
0045007	RoboColumn GigaCap DEAE-650M	0.2 mL*8 cols
0045008	RoboColumn GigaCap DEAE-650M	0.6 mL*8 cols
0045011	RoboColumn Q-600C AR	0.2 mL*8 cols
0045012	RoboColumn Q-600C AR	0.6 mL*8 cols
0045021	RoboColumn NH <sub>2</sub> -750F	0.2 mL*8 cols
0045022	RoboColumn NH <sub>2</sub> -750F	0.6 mL*8 cols

Part #	Description	Package description
<b>Mixed-Mode</b>		
0045051	RoboColumn MX-Trp-650M	0.2 mL*8 cols
0045052	RoboColumn MX-Trp-650M	0.6 mL*8 cols

**Hydrophobic Interaction**

0045031	RoboColumn Phenyl-600M	0.2 mL*8 cols
0045032	RoboColumn Phenyl-600M	0.6 mL*8 cols
0045033	RoboColumn Butyl-600M	0.2 mL*8 cols
0045034	RoboColumn Butyl-600M	0.6 mL*8 cols
0045035	RoboColumn PPG-600M	0.2 mL*8 cols
0045036	RoboColumn PPG-600M	0.6 mL*8 cols
0045037	RoboColumn Phenyl-650M	0.2 mL*8 cols
0045038	RoboColumn Phenyl-650M	0.6 mL*8 cols

**Affinity**

0045061	RoboColumn AF-rProtein A-650F	0.2 mL*8 cols
0045062	RoboColumn AF-rProtein A-650F	0.6 mL*8 cols
0045063	RoboColumn AF-rProtein A HC-650F	0.2 mL*8 cols
0045064	RoboColumn AF-rProtein A HC-650F	0.6 mL*8 cols



# PROCESS DEVELOPMENT

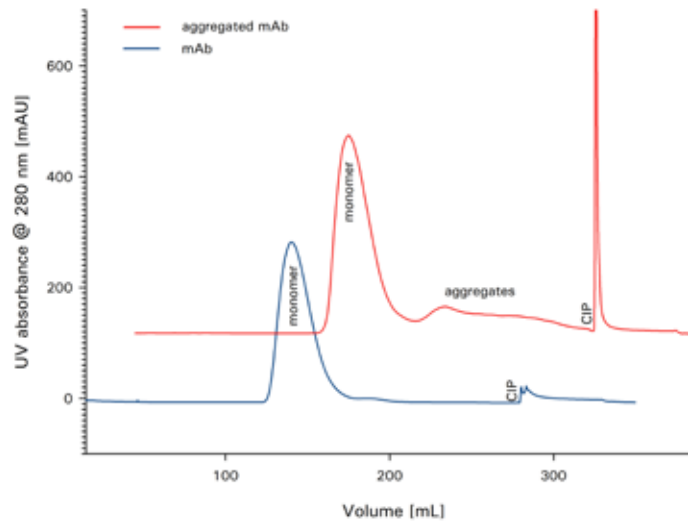
## MiniChrom for TOYOPEARL and TSKgel

TOYOPEARL and TSKgel media are now available in the well-known 5 mL MiniChrom format (8 mm ID x 100 mm) for parameter screening, method optimization and/or small scale purifications. The 5 mL MiniChrom columns are the ideal tools to further optimize the purification method and to confirm the operational window after having selected a resin for a certain purification task by resin screening, e.g. with ToyoScreen cartridges on conventional LC systems or by high throughput screening using RoboColumns on robotic workstations.

MiniChrom columns are made of biocompatible polyethylene and polypropylene. Each column is individually packed under optimum compression, ensuring consistent experimental results. The columns can be connected directly to any laboratory liquid chromatography system via standard connectors (M10-32 for 1/16" tubing) and are ready for equilibration in the buffer of choice. Two columns can be connected in series to increase the bed height in order to model real conditions in pilot scale or for scale-down experiments.

MiniChrom columns for TOYOPEARL and TSKgel are packed by Atoll GmbH and are available with a broad range of ion exchange, hydrophobic interaction, mixed-mode, and Protein A affinity resins. Figure 1 shows the mixed-mode separation of a monoclonal antibody and its aggregates on a 5 mL MiniChrom MX-Trp-650M column.

**FIGURE 6** Mixed-mode separation on MiniChrom MX-Trp-650M



Column: MiniChrom MX-Trp-650M, 8 mm ID x 10 cm L, 5 mL  
 Mobile phase: A: 100 mmol/L acetate buffer (pH 4.3) + 200 mmol/L NaCl, B: 100 mmol/L acetate buffer (pH 5.6) + 500 mmol/L NaCl;  
 Flow rate: 150 cm/h; Gradient: 5 CV 100% A, 50 CV linear gradient from 100% A to 100% B  
 Sample: 5 mL monoclonal antibody 5 mg/mL  
 5 mL aggregated monoclonal antibody (1 h, pH 2.7 @ RT) 5 mg/L

### FEATURES

- Pre-packed with TOYOPEARL or TSKgel media
- Common column format (5 mL, 8 x 100 mm)
- Reliable packing quality

### BENEFITS

- Ready to use with any LC system
- Ideal for method optimization and small scale purifications
- Reproducible results






**➤ ORDERING INFORMATION**

<i>Part #</i>	<i>Description</i>	<i>Package description</i>	<i>Part #</i>	<i>Description</i>	<i>Package description</i>
<b>MiniChrom Columns</b>			<b>Hydrophobic Interaction</b>		
<b>Ion Exchange</b>			<b>Mixed Mode</b>		
0045108	MiniChrom TOYOPEARL NH <sub>2</sub> -750F, 5 mL	8 mm ID x 100 mm L	0045121	MiniChrom TOYOPEARL Phenyl-650M, 5 mL	8 mm ID x 100 mm L
0045101	MiniChrom TOYOPEARL GigaCap S-650M, 5 mL	8 mm ID x 100 mm L	0045122	MiniChrom TOYOPEARL Phenyl-650S, 5 mL	8 mm ID x 100 mm L
0045102	MiniChrom TOYOPEARL GigaCap S-650S, 5 mL	8 mm ID x 100 mm L	0045123	MiniChrom TOYOPEARL Phenyl-600M, 5 mL	8 mm ID x 100 mm L
0045103	MiniChrom TOYOPEARL GigaCap CM-650M, 5 mL	8 mm ID x 100 mm L	0045124	MiniChrom TOYOPEARL PPG-600M, 5 mL	8 mm ID x 100 mm L
0045104	MiniChrom TOYOPEARL GigaCap Q-650M, 5 mL	8 mm ID x 100 mm L	0045125	MiniChrom TOYOPEARL Butyl-650M, 5 mL	8 mm ID x 100 mm L
0045105	MiniChrom TOYOPEARL GigaCap Q-650S, 5 mL	8 mm ID x 100 mm L	0045126	MiniChrom TOYOPEARL Butyl-650S, 5 mL	8 mm ID x 100 mm L
0045106	MiniChrom TOYOPEARL GigaCap DEAE-650M, 5 mL	8 mm ID x 100 mm L	0045127	MiniChrom TOYOPEARL Butyl-600M, 5 mL	8 mm ID x 100 mm L
0045107	MiniChrom TSKgel SuperQ-5PW (20), 5 mL	8 mm ID x 100 mm L	<b>Affinity</b>		
0045109	MiniChrom TOYOPEARL Super Q-650M	8 mm ID x 100 mm L	0045151	MiniChrom TOYOPEARL MX-Trp-650M 5 mL	8 mm ID x 100 mm L
0045110	MiniChrom TOYOPEARL SP-650M	8 mm ID x 100 mm L	0045161	MiniChrom TOYOPEARL AF-rProtein A HC-650M 5 mL	8 mm ID x 100 mm L
0045111	MiniChrom TOYOPEARL SP-650S	8 mm ID x 100 mm L			
0045112	MiniChrom TOYOPEARL DEAE-650M	8 mm ID x 100 mm L			

# PROCESS DEVELOPMENT

## TOYOPEARL AND TSKgel LABPAK MEDIA

TOYOPEARL and TSKgel LabPak media products are small package sizes of TOYOPEARL and TSKgel bulk media products. Typically they contain three or four different ligand types offered for a particular chromatography mode.

They are useful for developmental scientists and engineers who wish to familiarize themselves with the physical properties of resins in different buffer systems:

- slurry and reslurry mechanics
- resin handling during column packing
- mechanical strength relative to other resin backbones
- degree of compressibility

The larger resin amounts in LabPak products allow the packing of wider bore and longer columns than available in the ToyoScreen products. This helps the developmental scientist or engineer to more accurately determine the resin's:

- dynamic binding capacity
- selectivity
- column efficiency
- operating conditions

### ➤ ORDERING INFORMATION

Part #	Description	Container size	Part #	Description	Container size
<b>TSKgel LABPAKS</b>			<b>TOYOPEARL LABPAKS</b>		
<b>Ion Exchange</b>			<b>Size Exclusion</b>		
0043380	IEXPAK PW, 20 µm (DEAE-5PW, SP-5PW, SuperQ-5PW)	3 x 25 mL	0019820	SECPAK HP, 30 µm (HW-40, 50, 55, 65S)	4 x 150 mL
0043280	IEXPAK PW, 30 µm (DEAE-5PW, SP-5PW, SuperQ-5PW)	3 x 25 mL	0019821	SECPAK LMW, 45 µm (HW-40, 50, 55F)	3 x 150 mL
<b>Hydrophobic Interaction</b>			0019819	SECPAK HMW, 45 µm (HW-55, 65, 75F)	3 x 150 mL
0043278	HICPAK PW, 20 µm (Ether-5PW, Phenyl-5PW)	2 x 25 mL	<b>Ion Exchange</b>		
0043175	HICPAK PW, 30 µm (Ether-5PW, Phenyl-5PW)	2 x 25 mL	0019817	IEXPAK HP, 35 µm (DEAE-650S, SP-650S, CM-650S, SuperQ-650S)	4 x 25 mL
			0043210	AIEXPAK, 75/100 µm (GigaCap Q-650M, SuperQ-650M, Q-600C AR)	3 x 100 mL
			0043220	CIEXPAK, 75/100 µm (GigaCap CM-650M/ S-650M, SP-550C)	3 x 100 mL
			<b>Hydrophobic Interaction</b>		
			0043150	HICPAK HP, 35 µm (Ether, Phenyl, Butyl-650S)	3 x 25 mL
			0019806	HICPAK, 65 µm (Ether, Phenyl, Butyl-650M)	3 x 25 mL
			0043125	HICPAK-C, 100 µm (Phenyl, Butyl, Hexyl-650C)	3 x 25 mL
			<b>Affinity</b>		
			0043400	AFFIPAK ACT, 65 µm (AF-Epoxy, Tresyl-650M)	2 x 5 g*
			0043410	AFFIPAK, 65 µm (AF-Amino, Carboxyl, Formyl-650 M)	3 x 10 mL

\*1 g is approximately 3.5 mL



## INTRODUCTION TO BULK RESINS FOR LABORATORY PURIFICATION

Tosoh Bioscience offers TOYOPEARL and TSKgel resins (media) in bulk quantities for laboratory-scale applications.

Although the resins can be applied to the purification of small as well as large MW compounds, TOYOPEARL and TSKgel resins are most useful for the separation of peptides, proteins, and oligonucleotides.

The focus of this section is on the use of bulk resins in laboratory applications. Please request the Process Chromatography Catalog for information about the use of TOYOPEARL and TSKgel for larger scale separations or visit our website at: [www.tosohbioscience.de](http://www.tosohbioscience.de).

### TOYOPEARL BULK RESIN

TOYOPEARL resins are hydrophilic, macroporous media for medium pressure liquid chromatographic applications.

The polymethacrylate backbone structure of TOYOPEARL packings assure excellent pressure/flow characteristics. TOYOPEARL is mechanically stable up to 0.3 MPa, which simplifies column packing by reducing the setup time and improving reproducibility from column to column.

The media is stable over the range of pH 2-12 for normal operating conditions and pH 1-13 for cleaning conditions. In most modes, TOYOPEARL is available in three grades, S (superfine) for highest performance, F (fine) and M (medium) for economical purification, and C (coarse) and EC (extra coarse) for capture. Consult **TABLE I** for particle sizes associated with the various chemistries and pore sizes.

### FEATURES

- chemistries available in Size Exclusion, Ion Exchange, Mixed-Mode, Hydrophobic Interaction and Affinity chromatography
- methacrylate backbone has hydrophilic surface properties
- TSKgel and TOYOPEARL bulk resin product lines feature the same ligand and backbone chemistries from 20 µm to 150 µm particle sizes
- SEC product line available in 5 pore sizes
- IEC, HIC and AFC products are based on 100, 75 and 50 nm pore size particles.
- chemical stability
- thermal stability
- mechanical stability
- column bed stability

### BENEFITS

- added flexibility during method development
- less non-specific adsorption
- high recovery of proteins, enzymes, glycoproteins
- simplified scale up from laboratory separation to process
- suitable for fractionation of large and small biopolymers
- high capacity and efficient chromatography of small protein and large biopolymers due to unrestricted access of available surface area
- cleanable resins in strong base or acid (pH 1-13)
- compatible with all water soluble organic solvents
- stable in chaotropic agents such as: guanidine hydrochloride, sodium dodecyl sulfate and urea
- autoclavable at 120°C
- wide range of operating temperature (4-60°C)
- linear relationship between flow rate and pressure drop
- constant bed volume over a wide range of salt concentrations

# PROCESS DEVELOPMENT

## PROCESS DEVELOPMENT BULK MEDIA

TOYOPEARL HW-type resins, available in pore sizes ranging from 5 nm to >100 nm, are employed in size exclusion chromatography (SEC). TOYOPEARL HW-65 and HW-55 resins are used as starting materials for the production of all other functionalized TOYOPEARL resins. The large pore size of HW-65 (100 nm) allows unhindered access of large proteins to the stationary phase, resulting in faster separation and shorter recycling times.

For predictable results during scale up, TOYOPEARL resins are based on the same chemistry as the prepacked TSKgel columns. This allows for seamless scale up from the laboratory to manufacturing.

### TSKgel BULK RESINS

TSKgel resins are larger particle size versions of the chemically equivalent methacrylic packing of analytical-scale TSKgel columns used for protein analysis and purification. The TSKgel resin product line consists of DEAE-5PW, SuperQ-5PW, SP-5PW, and SP-3PW resins for ion exchange, Tresyl-5PW resins for affinity chromatography, and Ether-5PW and Phenyl-5PW resins for HIC. TSKgel resins are often employed to simplify scale-up from analytical columns, as only the particle size is different. Their small particle sizes, high degree of cross-linking and high mechanical stability make TSKgel resins the preferred choice for high efficiency purifications.

TABLE I

Characteristics of TOYOPEARL and TSKgel media

Mode	Resin	Grade/particle size (µm)	Pore size (nm)**	MW range Proteins (Da)	Operating pH range	Max. pressure (MPa)
SEC	TOYOPEARL HW-40	S (20-40), F (30-60), C (50-100)	5	1 x 10 <sup>2</sup> - 1 x 10 <sup>4</sup>	2-12	0.3
	TOYOPEARL HW-50	S (20-40), F (30-60)	12.5	5 x 10 <sup>2</sup> - 8 x 10 <sup>4</sup>	2-12	0.3
	TOYOPEARL HW-55	S (20-40), F (30-60)	50	1 x 10 <sup>3</sup> - 7 x 10 <sup>5</sup>	2-12	0.3
	TOYOPEARL HW-65	S (20-40), F (30-60)	100	4 x 10 <sup>4</sup> - 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL HW-75	S (20-40), F (30-60)	> 100	5 x 10 <sup>5</sup> - 5 x 10 <sup>7</sup>	2-12	0.3
IEC	TSKgel SuperQ-5PW	20 and 30	100	< 5 x 10 <sup>6</sup>	2-12	2.0
	TSKgel DEAE-5PW	20 and 30	100	< 5 x 10 <sup>6</sup>	2-12	2.0
	TSKgel SP-5PW	20 and 30	100	< 5 x 10 <sup>6</sup>	2-12	2.0
	TSKgel SP-3PW	30	25	< 1 x 10 <sup>4</sup>	2-12	2.0
	TOYOPEARL SuperQ-650	S (20-50), M (40-90), C (50-150)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL DEAE-650	S (20-50), M (40-90), C (50-150)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL GigaCap Q-650	S (20-50), M (50-100)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL GigaCap DEAE-650	M (50-100)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL SP-650	S (20-50), M (40-90), C (50-150)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL CM-650	S (20-50), M (40-90), C (50-150)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL GigaCap S-650	S (20-50), M (50-100)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL GigaCap CM-650	M (50-100)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL QAE-550	C (50-150)	50	< 5 x 10 <sup>5</sup>	2-12	0.3
	TOYOPEARL Q-600C AR	C (50-150)	75	< 2.5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL NH <sub>2</sub> -750	F (30-60)	>1000	< 5 x 10 <sup>7</sup>	2-12	0.3
	TOYOPEARL SP-550	C (50-150)	50	< 5 x 10 <sup>5</sup>	2-12	0.3
	TOYOPEARL MegaCap II SP-550	EC (100-300)	50	< 5 x 10 <sup>5</sup>	2-12	0.3
MMC	TOYOPEARL MX-Trp-650M	M (50-100)	100	< 5 x 10 <sup>6</sup>	2-12	2.0
HIC	TSKgel Ether-5PW	20 and 30	100	< 5 x 10 <sup>6</sup>	2-12	2.0
	TSKgel Phenyl-5PW	20 and 30	100	< 5 x 10 <sup>6</sup>	2-12	2.0
	TOYOPEARL Ether-650	S (20-50), M (40-90)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL PPG-600	M (40-90)	75	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL Phenyl-600	M (40-90)	75	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL Butyl-600	M (40-90)	75	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL Phenyl-650	S (20-50), M (40-90), C (50-150)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL Butyl-650	S (20-50), M (40-90), C (50-150)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
	TOYOPEARL Super Butyl-550	C (50-150)	50	< 5 x 10 <sup>5</sup>	2-12	0.3
	TOYOPEARL Hexyl-650	C (50-150)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
	AFC	TSKgel Tresyl-5PW	10	100	< 5 x 10 <sup>6</sup>	2-12
TOYOPEARL AF-Chelate-650		M (40-90)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
TOYOPEARL AF-rProtein A HC-650		F (30-60)	100	< 5 x 10 <sup>6</sup>	N/A	0.3
TOYOPEARL AF-rProtein A		F (30-60)	100	< 5 x 10 <sup>6</sup>	N/A	0.3
TOYOPEARL AF-Tresyl-650		M (40-90)	100	< 5 x 10 <sup>6</sup>	N/A	0.3
TOYOPEARL AF-Epoxy-650		M (40-90)	100	< 5 x 10 <sup>6</sup>	N/A	0.3
TOYOPEARL AF-Formyl-650		M (40-90)	100	< 5 x 10 <sup>6</sup>	6-9	0.3
TOYOPEARL AF-Amino-650		M (40-90)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
TOYOPEARL AF-Carboxy-650		M (40-90)	100	< 5 x 10 <sup>6</sup>	2-12	0.3
TOYOPEARL AF-Red-650		M (40-90)	100	< 5 x 10 <sup>6</sup>	4-9	0.3
TOYOPEARL AF-Blue HC-650		M (40-90)	100	< 5 x 10 <sup>6</sup>	4-9	0.3
TOYOPEARL AF-Heparin HC-650		M (40-90)	100	< 5 x 10 <sup>6</sup>	5-10	0.3

\*\* nominal values; Pore size of base matrix



## TOYOPEARL BULK RESINS FOR SEC

### HIGHLIGHTS

- Pore sizes ranging from 5 nm to >100 nm
- Three particle sizes (S, F, C)
- HW-40 is ideal for desalting applications
- Easy to pack in semi-preparative and process scale columns

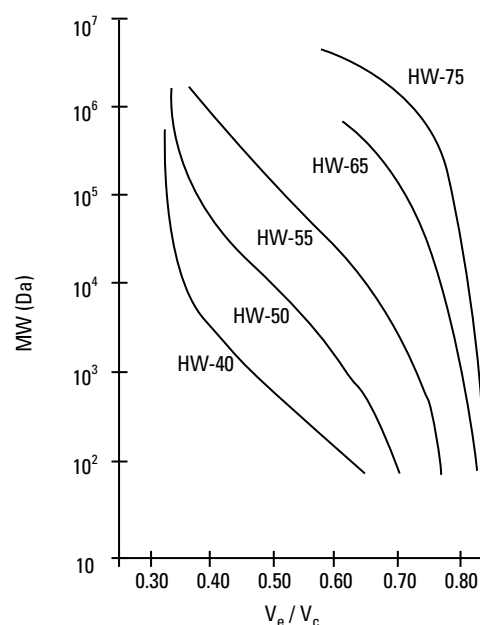
Size exclusion chromatography (SEC) is a common technique for separating molecules based on their apparent molecular size. For nearly twenty-five years, TOYOPEARL SEC bulk resins, with their macroporous packings, have been used for laboratory and production-scale biochromatography.

TOYOPEARL SEC resins are semi-rigid, spherical polymethacrylate beads. The resins have hydrophilic surfaces due to the presence of ether and hydroxyl groups. The numerous surface hydroxyl groups provide attachment points for other functional groups and ligands. **TABLE II** provides an overview of the TOYOPEARL SEC resin product line including corresponding molecular weight ranges of common target samples. Calibration curves of the TOYOPEARL HW-type resins determined with globular proteins are presented in **FIGURE 5**.

Ordering information for quantities <1 L is provided at the end of this section. For larger quantities, please contact customer service at **+49 (0) 6155 70437-30**. LABPAK kits are also available in popular combinations of TOYOPEARL media. See the page 99 for additional information.

**Applications:** proteins, peptides, amino acids, nucleic acids, and small molecular weight molecules. Please visit our website: [www.tosohbioscience.de](http://www.tosohbioscience.de) for extensive data on applications.

**FIGURE 5**  
Calibration curves for globular proteins on TOYOPEARL HW-type resins



Column: 22 mm ID x 30 cm L; Sample: protein standards;  
Elution: 0.06 mol/L phosphate buffer, pH 7, in 0.06 mol/L KCl;  
Legend:  $V_e$ =elution volume,  $V_c$ =column volume

**TABLE II**

Properties and molecular weight separation ranges for TOYOPEARL HW-type resins  
(HW = Hydrophilic, water-compatible polymeric base resins)

TOYOPEARL resin	Particle size ( $\mu\text{m}$ )	Pore size (nm)	Molecular weight of sample (Da)		
			PEG and PEO	Dextrans	Globular proteins
HW-40S	20 - 40	5	$1 \times 10^2 - 3 \times 10^3$	$1 \times 10^2 - 7 \times 10^3$	$1 \times 10^2 - 1 \times 10^4$
HW-40F	30 - 60	5			
HW-40C	50 - 100	5			
HW-50S	20 - 40	12.5	$1 \times 10^2 - 1.8 \times 10^4$	$5 \times 10^2 - 2 \times 10^4$	$5 \times 10^2 - 8 \times 10^4$
HW-50F	30 - 60	12.5			
HW-55S	20 - 40	50	$1 \times 10^2 - 1.5 \times 10^5$	$1 \times 10^3 - 2 \times 10^5$	$1 \times 10^3 - 7 \times 10^5$
HW-55F	30 - 60	50			
HW-65S	20 - 40	100	$5 \times 10^2 - 1 \times 10^6$	$1 \times 10^4 - 1 \times 10^6$	$4 \times 10^4 - 5 \times 10^6$
HW-65F	30 - 60	100			
HW-75F	30 - 60	>100	$4 \times 10^3 - 5 \times 10^6$	$1 \times 10^5 - 1 \times 10^7$	$5 \times 10^5 - 5 \times 10^7$

# PROCESS DEVELOPMENT

## TOYOPEARL AND TSKgel BULK RESINS FOR IEC

### HIGHLIGHTS

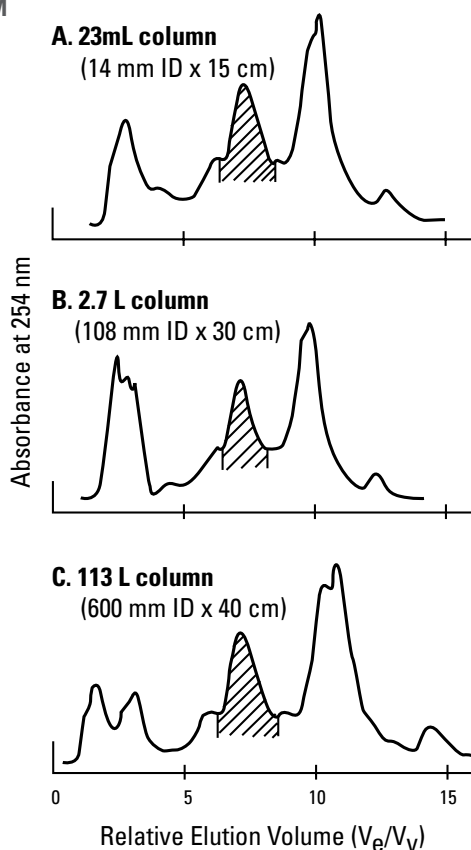
- TOYOPEARL GigaCap®S-650, CM-650M, DEAE-650, and Q-650 resins are high capacity ion exchange resins featuring high dynamic binding capacities for both small molecules like insulin and larger proteins like monoclonal antibodies.
- Salt tolerant TOYOPEARL NH<sub>2</sub>-750F anion exchanger with >100 nm pore size.
- Weak and strong anion and cation exchangers are offered in both product lines.
- Standard 100 nm pore size for large biopolymers and 50 nm pore size packing for optimal binding capacity are available.
- High efficiency TSKgel resins scale up directly from TSKgel analytical columns.

For separating mixtures of biomolecules, Ion Exchange Chromatography (IEC) is known for its high resolution and high capacity. It is very effective in the initial capture step of a chromatography process. IEC is also useful for further purification and/or polishing. It can complement other chromatographic techniques in the design of an economical downstream purification process. IEC is often used as a purification step before HIC, SEC, and RPC. IEC will also purify and concentrate the

target molecule in one step when the sample is diluted. This also allows it to be used as a concentration step after SEC. A 5000-fold scale-up of a  $\alpha$ -galactosidase enzyme purification was accomplished using TOYOPEARL DEAE-650M. The chromatograms in **FIGURE 6** demonstrate the excellent scale up characteristics of TOYOPEARL ion exchange media. Gradient slope and particle diameter remained unchanged. Linear velocity was reduced by 15% in the largest scale separation, and resolution actually improved relative to the smallest scale separation. This may be partly attributed to increased bed height and the slower linear velocity. Although the column volume was increased in part by increasing the bed height, the principal change in column volume was a result of the greater column diameter (1.4 cm to 60 cm L). This example illustrates how TOYOPEARL media can be conveniently scaled up from laboratory to production scale applications using the same particle size if desired.

Because the correct choice of an ion exchange resin can have a considerable impact on the economy of a process, Tosoh Bioscience provides many product options in both TOYOPEARL and TSKgel IEC bulk polymeric media. See **TABLE III** for a complete listing of available particle sizes. Ordering information for quantities < 1 L is provided at the end of this section.

**FIGURE 6**  
Process scale-up purification of  $\beta$ -galactosidase with TOYOPEARL DEAE-650M

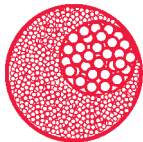


Column: TOYOPEARL DEAE-650 M; Sample: 1%  $\beta$ -galactosidase: A. 8 mL; B. 1L; C. 40L Elution: linear gradient from 0.03 to 0.10 mol/L NaCl in 0.014 mol/L Tris-HCl (pH7.7); Flow rate: A. 1.0 mL/min; B. 60 mL/min; C. 1.6 L/min; Linear velocity: A. 39 cm/h; B. 40 cm/h; C. 34 cm/h; Detection: UV@254nm

**TABLE III**  
TOYOPEARL and TSKgel Ion Exchange resins

Description	Type*	Part. size ( $\mu$ m)
<b>Anion Exchange</b>		
TSKgel DEAE-5PW	W	20, 30
TSKgel SuperQ-5PW	S	20, 30
TOYOPEARL DEAE-650	W	35, 65, 100
TOYOPEARL SuperQ-650	S	35, 65, 100
TOYOPEARL QAE-550	S	100
TOYOPEARL Q-600 AR	S	100
TOYOPEARL GigaCap Q-650M	S	35, 75
TOYOPEARL GigaCap DEAE-650M	W	75
TOYOPEARL NH <sub>2</sub> -750F	S	45
<b>Cation Exchange</b>		
TSKgel SP-5PW	S	20, 30
TSKgel SP-3PW	S	30
TOYOPEARL CM-650	W	35, 65, 100
TOYOPEARL GigaCap CM-650M	W	75
TOYOPEARL SP-550	S	100
TOYOPEARL SP-650	S	35, 65, 100
TOYOPEARL MegaCap II SP-550EC	S	100-300
TOYOPEARL GigaCap S-650M	S	35, 75

\*W = Weak; S = Strong



## TOYOPEARL AND TSKgel BULK RESINS FOR MIXED-MODE CHROMATOGRAPHY

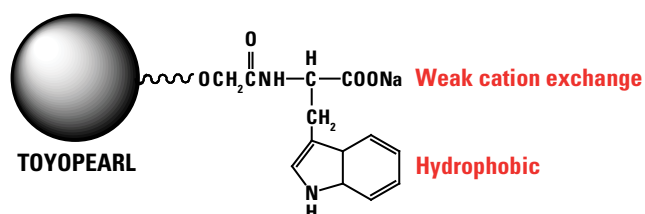
### HIGHLIGHTS

- TOYOPEARL MX-Trp-650M is a multimodal cation exchange resin
- It provides high binding capacity for IgG and other proteins
- It tolerates high conductivity feedstocks
- Target molecules elute under mild conditions in sharp peaks

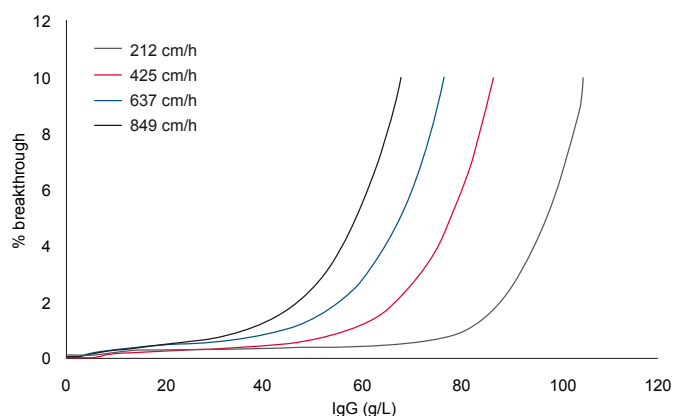
Multimodal or mixed-mode chromatography expands the range of chromatographic modes applied in biopurification. Mixed-mode media combine ionic and hydrophobic interactions and offer new selectivities and a higher salt tolerance than traditional ion exchange media. Mixed-mode media can be used for direct processing of clarified feedstocks at physiological salt concentrations as well as for intermediate and polishing applications. The salt tolerance of the recently introduced TOYOPEARL NH<sub>2</sub>-750F anion exchange resin is to a certain extent also based on mixed-mode interactions. Nevertheless, this resin is listed in the ion exchange section.

TOYOPEARL MX-Trp-650M is a multimodal cation exchange resin with unique selectivity and high recovery. It provides high protein binding capacities and tolerates high conductivity feedstocks. In addition to ionic groups its ligand also carries hydrophobic regions. Thus, the binding of target molecules is determined by electrostatic and hydrophobic contributions. TOYOPEARL MX-Trp-650M is especially suited for the purification of target molecules that are difficult to purify using common purification platforms.

➤ **FIGURE 7**  
TOYOPEARL MX-Trp-650M STRUCTURE



➤ **FIGURE 8**  
IgG binding capacity

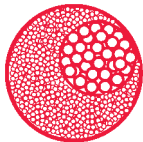


Column: TOYOPEARL MX-Trp-650M (6 mm ID x 4 cm);  
 Sample: polyclonal human IgG (1 mg/mL) in 0.05 mol/L NaAc + 0.1 mol/L sodium chloride (pH 4.7); Linear velocity: 212, 425, 637, 849 cm/h;  
 Detection: UV @ 280 nm



# PROCESS DEVELOPMENT





## TOYOPEARL AND TSKgel BULK RESINS FOR HIC

### HIGHLIGHTS

- A wide range of hydrophobicities is suitable for most proteins.
- Standard 100 nm pore size is available for large biopolymers, and three Butyl pore sizes (50 nm, 75 nm and 100 nm) are available.
- TOYOPEARL "600M" series of HIC resins with optimized pore size of 75 nm for antibody separation. Phenyl-600M and Butyl-600M with highest DBCs for IgG.
- Seamless scale up from high efficiency TSKgel 5PW-type analytical columns is possible.

Hydrophobic Interaction Chromatography (HIC) has become a popular mode of chromatography for the purification of biopolymers at analytical as well as preparative scale. This is accomplished by the interaction of hydrophobic ligands on the base matrix with the hydrophobic areas located on the surface of proteins. HIC is an excellent complement to size exclusion and ion exchange chromatography in difficult separations, particularly those where the contaminants are of similar pI or molecular weight. It is often preferred over reversed phase chromatography when preservation of biological activity of the protein is of utmost importance.

Tosoh Bioscience offers both the TSKgel and TOYOPEARL resin product lines for HIC. See **TABLE IV** for a complete listing of functionalities. Each product line has similar backbone chemistry. TSKgel 5PW-type resins possess a higher degree of cross-linking than the corresponding TOYOPEARL resins. Additionally, choices in particle size are offered to match the desired resolution and throughput. A variety of HIC bulk media are offered as LABPAK kits in quantities < 1 L and in a combination of resins with varying functionalities. Additionally, HIC media are available in ToyoScreen process development columns for convenient scouting and methods development.

Ordering information for quantities < 1 L is provided at the end of this section.

### TABLE IV TOYOPEARL and TSKgel HIC resins

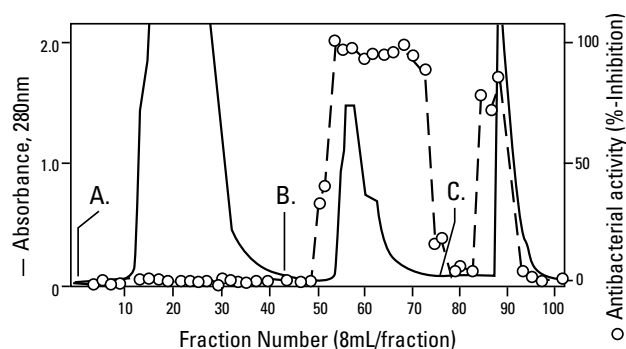
Description	Strength*	Part. size grades (µm)
TSKgel Ether-5PW	1	20, 30
TOYOPEARL Ether-650	1	35, 65
TOYOPEARL PPG-600	2	65, 100
TSKgel Phenyl-5PW	3	20, 30
TOYOPEARL Phenyl-650	3	35, 65, 100
TOYOPEARL Phenyl-600	4	65
TOYOPEARL Butyl-600	4	65
TOYOPEARL Butyl-650	4	35, 65, 100
TOYOPEARL SuperButyl-550	4	100
TOYOPEARL Hexyl-650	5	100

\* Relative scale: 1 = least hydrophobic, 5 = most hydrophobic.

### APPLICATIONS:

HIC resins can be applied to separate/purify proteins with similar chemical or structural properties, plasmids and monoclonal antibodies. See **FIGURE 9** for separation of large glycoprotein from crude extract on TOYOPEARL Butyl-650S. Please visit our website: [www.tosohbioscience.de](http://www.tosohbioscience.de) for extensive application data.

**FIGURE 9**  
Large glycoprotein purified on TOYOPEARL Butyl-650S



Column: TOYOPEARL Butyl-650S, 22 mm ID x 26 cm L;  
Sample: crude protein from sea hare *Aplysia kurodai*;  
Elution: multi-step (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> in 50 mmol/L phosphate buffer, pH 7.0  
A. load & wash: 40 % saturated (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>  
B. 20% saturated (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>  
C. 0% saturated (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>

# PROCESS DEVELOPMENT

## TOYOPEARL RESINS FOR AFC

### HIGHLIGHTS

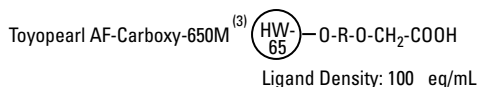
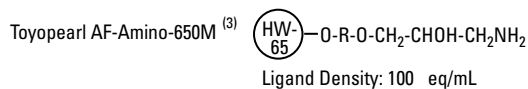
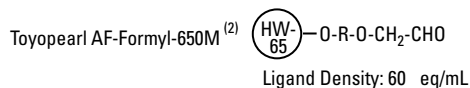
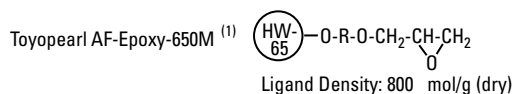
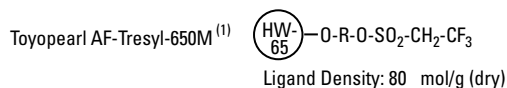
- New AF-rProtein A-HC 650F resin for antibody purification.
- Active, reactive and group specific resins
- Provided in standard 100 nm pore size for high capacity of large biopolymers.
- TOYOPEARL AF-Blue HC-650M is available for albumin and interferon applications with the lowest leaching blue.
- TOYOPEARL AF-Heparin HC-650M high capacity resin exhibits an Antithrombin III dynamic capacity of 4 mg/mL.

TOYOPEARL media for Affinity Chromatography (AFC) are based on TOYOPEARL HW-65 resin and functionalized with either chemically active groups or group-specific ligands. Resins with activated functional groups are ready for direct coupling of a protein or other ligand, while resins with reactive groups employ coupling or reductive amination to achieve covalent bonding. The 100 nm pore size common to all TOYOPEARL affinity resins accommodates proteins up to 5,000,000 Da.

In general, TOYOPEARL AF-Tresyl-650M and AF-Formyl-650M are recommended for coupling proteins, while AF-Epoxy-650M is suited for coupling low molecular weight ligands. TOYOPEARL AF-Amino-650M and TOYOPEARL AF-Carboxy-650M may be used in either application. TOYOPEARL AF-Heparin HC-650M interacts with a wide range of biomolecules including plasma components, lipoprotein lipase, collagenase, and DNA polymerase. The structures of TOYOPEARL activated and reactive ligands are given in FIGURE 10, while the structures of TOYOPEARL group-specific ligands are listed in FIGURE 11.

### ➤ FIGURE 10

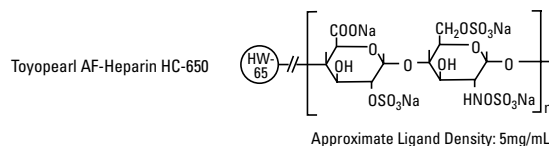
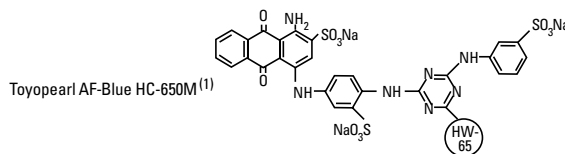
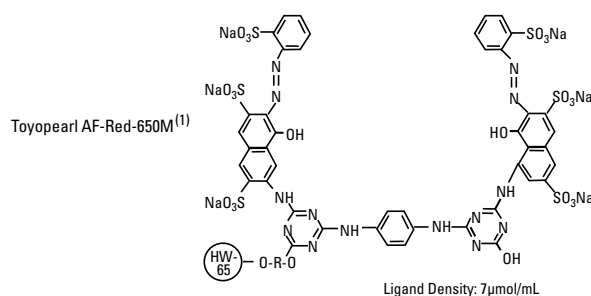
#### Activated and reactive TOYOPEARL affinity resins



(1) Provided as dry, free-flowing powder.  
One gram of dry powder produces about 3.5 mL of hydrated resin.  
(2) Provided as aqueous slurry, containing 1% glutaraldehyde.  
(3) Provided as aqueous slurry, containing 20% ethanol.

TOYOPEARL AF-rProtein A HC-650F designed for efficient and robust purification of antibodies. The newly developed recombinant protein A ligands are derived from one of the IgG-binding domains of the staphylococcus aureus protein A (FIGURE 12). TOYOPEARL AF-rProtein A HC-650F binds immunoglobulin G with high binding capacity and at high flow rates. This reduces column and buffer volumes and allows fast loading procedures.

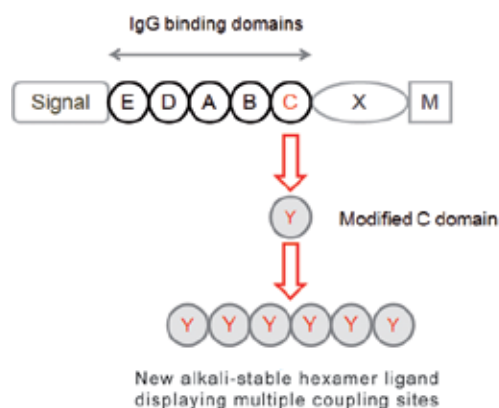
### ➤ FIGURE 11 Group-specific TOYOPEARL affinity resins



(1) Provided as an aqueous slurry containing 20% ethanol, v/v in 1mol/L NaCl.  
(2) Provided as an aqueous slurry containing 20% ethanol.

### ➤ FIGURE 12

#### Recombinant Protein A derived ligand





## ► ORDERING INFORMATION

Part #	Description	Container size	Part #	Description	Container size
<b>A. Size Exclusion Chromatography</b>					
<b>TOYOPEARL bulk resins</b>					
0019809	HW-40S, 30 µm	150 mL	0019804	DEAE-650S, 35 µm	25 mL
0007451	HW-40S, 30 µm	250 mL	0007472	DEAE-650S, 35 µm	250 mL
0019808	HW-40F, 45 µm	150 mL	0043201	DEAE-650M, 65 µm	100 mL
0007448	HW-40F, 45 µm	500 mL	0007473	DEAE-650M, 65 µm	250 mL
0019807	HW-40C, 75 µm	150 mL	0007988	DEAE-650C, 100 µm	250 mL
0007449	HW-40C, 75 µm	500 mL	0022865	GigaCap DEAE-650M, 75 µm	100 mL
0019811	HW-50S, 30µm	150 mL	0022866	GigaCap DEAE-650M, 75 µm	250 mL
0007455	HW-50S, 30µm	250 mL	0022881	GigaCap Q-650S, 35 µm	25 mL
0019810	HW-50F, 45 µm	150 mL	0022882	GigaCap Q-650S, 35 µm	250 mL
0007453	HW-50F, 45 µm	500 mL	0021854	GigaCap Q-650M, 75 µm	100 mL
0019813	HW-55S, 30 µm	150 mL	0021855	GigaCap Q-650M, 75 µm	250 mL
0007459	HW-55S, 30 µm	250 mL	0023438	NH <sub>2</sub> -750F, 45 µm	100 mL
0019812	HW-55F, 45 µm	150 mL	0023439	NH <sub>2</sub> -750F, 45 µm	250 mL
0007457	HW-55F, 45 µm	500 mL	<b>C. Cation Exchange Chromatography</b>		
0019815	HW-65S, 30 µm	150 mL	<b>TSKgel bulk resins</b>		
0007467	HW-65S, 30 µm	250 mL	0043382	SP-5PW (20)	25 mL
0019814	HW-65F, 45 µm	150 mL	0014714	SP-5PW (20)	250 mL
0007465	HW-65F, 45 µm	500 mL	0043282	SP-5PW (30)	25 mL
0021481	HW-65C, 75 µm	150mL	0014716	SP-5PW (30)	250 mL
0007466	HW-65C, 75 µm	500mL	0021976	SP-3PW (30)	25 mL
0019816	HW-75F, 45 µm	150 mL	0021977	SP-3PW (30)	250 mL
0007469	HW-75F, 45 µm	500 mL	<b>TOYOPEARL bulk resins</b>		
<b>B. Anion Exchange Chromatography</b>					
<b>TSKgel bulk resins</b>					
0043381	DEAE-5PW (20)	25 mL	0019803	CM-650S, 35 µm	25 mL
0014710	DEAE-5PW (20)	250 mL	0007474	CM-650S, 35 µm	250 mL
0043281	DEAE-5PW (30)	25 mL	0043203	CM-650M, 65 µm	100 mL
0014712	DEAE-5PW (30)	250 mL	0007475	CM-650M, 65 µm	250 mL
0043383	SuperQ-5PW (20)	25 mL	0007991	CM-650C, 100 µm	250 mL
0018535	SuperQ-5PW (20)	250 mL	0019822	SP-650S, 35 µm	25 mL
0043283	SuperQ-5PW (30)	25 mL	0008437	SP-650S, 35 µm	250 mL
0018536	SuperQ-5PW (30)	250 mL	0043202	SP-650M, 65 µm	100 mL
<b>TOYOPEARL bulk resins</b>					
0019823	SuperQ-650S, 35 µm	25 mL	0007997	SP-650M, 65 µm	250 mL
0017223	SuperQ-650S, 35 µm	250 mL	0007994	SP-650C, 100 µm	250 mL
0043205	SuperQ-650M, 65 µm	100 mL	0043272	SP-550C, 100 µm	100 mL
0017227	SuperQ-650M, 65 µm	250 mL	0014028	SP-550C, 100 µm	250 mL
0043275	SuperQ-650C, 100 µm	100 mL	0021804	MegaCap II SP-550EC, 160 µm	100 mL
0017231	SuperQ-650C, 100 µm	250 mL	0021805	MegaCap II SP-550EC, 160 µm	250 mL
0043271	QAE-550C, 100 µm	100 mL	0022875	GigaCap S-650S, 35 µm	25 mL
0014026	QAE-550C, 100 µm	250 mL	0022876	GigaCap S-650S, 35 µm	250 mL
0021985	Q-600C AR, 100 µm	100 mL	0021833	GigaCap S-650M, 75 µm	100 mL
0021986	Q-600C AR, 100 µm -	250 mL	0021834	GigaCap S-650M, 75 µm	250 mL
<b>D. Mixed-Mode</b>					
<b>TOYOPEARL bulk resins</b>					
0022817	MX-Trp-650M, 75 µm	25 mL	0021946	GigaCap CM-650M, 75 µm	100 mL
0022818	MX-Trp--650M, 75 µm	100 mL	0021947	GigaCap CM-650M, 75 µm	250 mL

# PROCESS DEVELOPMENT

## BULK RESINS

➤ ORDERING INFORMATION

Part #	Description	Container size	Part #	Description	Container size
<b>E. Hydrophobic Interaction Chromatography</b>			<b>F. Affinity Chromatography</b>		
<b>TSKgel bulk resins</b>			<b>TSKgel bulk resins</b>		
0043276	Ether-5PW (20)	25 mL	0016208	Tresyl-5PW (10)	2 g*
0016052	Ether-5PW (20)	250 mL	<b>TOYOPEARL bulk resins</b>		
0043176	Ether-5PW (30)	25 mL	0023425	AF-rProtein A HC-650F, 45 µm	10 mL
0016050	Ether-5PW (30)	250 mL	0023426	AF-rProtein A HC-650F, 45 µm	25 mL
0043277	Phenyl-5PW (20)	25 mL	0023427	AF-rProtein A HC-650F, 45 µm	100 mL
0014718	Phenyl-5PW (20)	250 mL	0022803	AF-rProtein A-650F, 45 µm	10 mL
0043177	Phenyl-5PW (30)	25 mL	0022804	AF-rProtein A-650F, 45 µm	25 mL
0014720	Phenyl-5PW (30)	250 mL	0022805	AF-rProtein A-650F, 45 µm	100 mL
<b>TOYOPEARL bulk resins</b>			0043411	AF-Amino-650M, 65 µm	10 mL
0019955	SuperButyl-550C, 100 µm	25 mL	0008002	AF-Amino-650M, 65 µm	25 mL
0019956	SuperButyl-550C, 100 µm	100 mL	0008039	AF-Amino-650M, 65 µm	100 mL
0021448	Butyl-600M, 65 µm	25 mL	0019688	AF-Blue HC-650M, 65 µm	25 mL
0021449	Butyl-600M, 65 µm	100 mL	0019689	AF-Blue HC-650M, 65 µm	100 mL
0043153	Butyl-650S, 35 µm	25 mL	0043412	AF-Carboxy-650M, 65 µm	10 mL
0007476	Butyl-650S, 35 µm	100 mL	0008006	AF-Carboxy-650M, 65 µm	25 mL
0019802	Butyl-650M, 65 µm	25 mL	0008041	AF-Carboxy-650M, 65 µm	100 mL
0007477	Butyl-650M, 65 µm	100 mL	0014475	AF-Chelate-650M, 65 µm	25 mL
0043127	Butyl-650C, 100 µm	25 mL	0019800	AF-Chelate-650M, 65 µm	100 mL
0007478	Butyl-650C, 100 µm	100 mL	0043402	AF-Epoxy-650M, 65 µm	5 g*
0043151	Ether-650S, 35 µm	25 mL	0008000	AF-Epoxy-650M, 65 µm	10 g*
0016172	Ether-650S, 35 µm	100 mL	0008038	AF-Epoxy-650M, 65 µm	100 g*
0019805	Ether-650M, 65 µm	25 mL	0043413	AF-Formyl-650M, 65 µm	10 mL
0016173	Ether-650M, 65 µm	100 mL	0008004	AF-Formyl-650M, 65 µm	25 mL
0044465	Hexyl-650C, 100 µm	25 mL	0008040	AF-Formyl-650M, 65 µm	100 mL
0019026	Hexyl-650C, 100 µm	100 mL	0020030	AF-Heparin-HC-650M, 65 µm	10 mL
0021887	Phenyl-600M, 65 µm	25 mL	0020031	AF-Heparin-HC-650M, 65 µm	100 mL
0021888	Phenyl-600M, 65 µm	100 mL	0008651	AF-Red-650M, 65 µm	25 mL
0043152	Phenyl-650S, 35 µm	25 mL	0019801	AF-Red-650M, 65 µm	100 mL
0014477	Phenyl-650S, 35 µm	100 mL	0014471	AF-Tresyl-650M, 65 µm	5 g*
0019818	Phenyl-650M, 65 µm	25 mL	0014472	AF-Tresyl-650M, 65 µm	100 g*
0014478	Phenyl-650M, 65 µm	100 mL	*1 g is approximately 3.5 mL		
0043126	Phenyl-650C, 100 µm	25 mL			
0014479	Phenyl-650C, 100 µm	100 mL			
0021301	PPG-600M, 65 µm	25 mL			
0021302	PPG-600M, 65 µm	100 mL			

