

Technical note

J.T. Baker® BAKERBOND® PolyCSX™ multimode strong cation exchange chromatography resin

INTRODUCTION

Utilizing a unique surface chemistry that combines strong cation exchange functionality with weak cationic and weak anionic exchange sites, BAKERBOND® PolyCSX™ multimode cation exchange chromatography media provides unique selectivity to enable higher purification performance across a wide range of biopharmaceuticals.

Designed and manufactured by Avantor to the high standards established by our J.T. Baker® brand, the BAKERBOND® PolyCSX™ resin provides better selectivity than conventional strong cation exchange media that enables separation of proteins and peptides having similar isoelectric points (pI). Its fully porous and crosslinked methacrylate spherical beads provide excellent mechanical and chemical stability and are optimized for maximum capacity, high mass transfer, and relatively low backpressure. The resin is provided in a non-hazardous, non-flammable storage solution through Avantor's global supply chain.

Used with Avantor's proven J.T. Baker® family of process chromatography buffers and additives, the BAKERBOND® PolyCSX™ resin can deliver greater efficiencies and higher purity profiles to biopharma chromatography schemes.

FEATURES

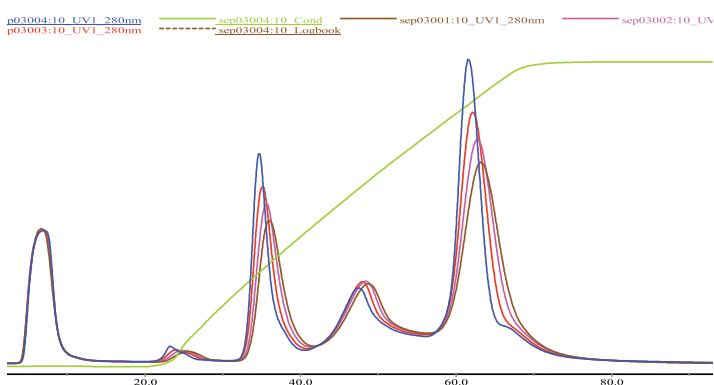
- Unique selectivity delivered by its proprietary multimode functionality
- High ionic capacity to improve separation and enable operation at high salt concentration
- Spherical polymethacrylate beads enable uniform packing and provide mechanical stability and chemical resistance for ease-of-use
- Delivery in a non-hazardous, non-flammable buffer solution to eliminate burdensome handling requirements.



UNIQUE SELECTIVITY

BAKERBOND® PolyCSX™ cation exchange resin is inherently well suited to discriminate between extremely similar molecules. Its unique surface chemistry is able to discern minute differences in primary structure resulting in a unique selectivity, improved separation performance, and higher efficiency downstream operations. As illustrated in Fig. 1, molecules with similar pI such as ovalbumin, hemoglobin, and lactoferrin are well resolved and can be easily separated.

Fig. 1. Proteins of similar isoelectric points (pI) separated at high flow rates



The x-axis represents volume instead of time and no loss in resolution is evident.

Sample size: 3.6 mL.

Sample: 1.5 mg/mL ovalbumin, 0.9 mg/mL IgG, 1.8 mg/mL hemoglobin, 1.8 mg/mL lactoferrin in binding buffer.

Column: 7.75 × 100 mm packed with PolyCSX.

Binding buffer: 137 mM HAc/TEA 32.9, mM NaCl pH 4.7.

Elution buffer: 1.0 M NaCl in Bind.

Buffer, Gradient: 0 to 100% elution buffer in 10 CV.

Flow rate: 204 cm/hr, 408 cm/hr, 611 m/hr and 815 cm/hr.

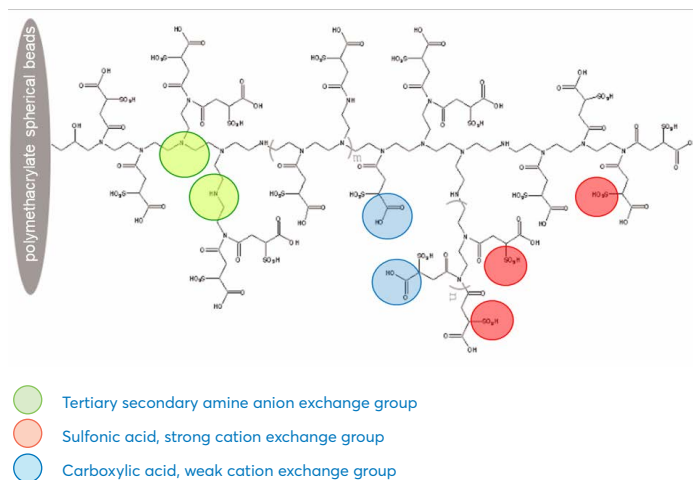
Blue: 1.6 mL/min (204 cm/hr)

Red: 3.2 mL/min (408 cm/hr)

Pink: 4.8 mL/min (611 cm/hr)

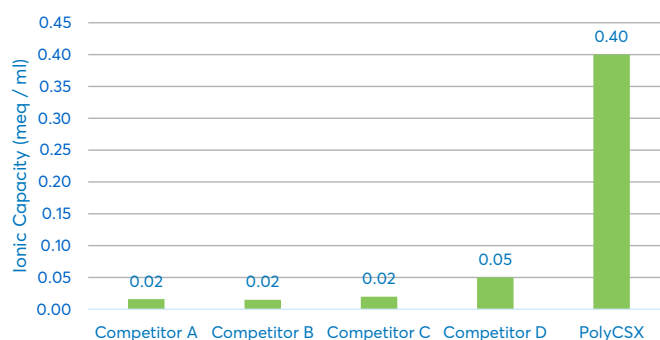
Brown: 6.4 mL/min (815 cm/hr).

The ability to discern these minute molecular differences is driven by the resin's multimode functionality. The multimode functionality of PolyCSX™ is obtained by covalently bonding PEI to the surface of highly cross linked polymethacrylate beads and functionalizing PEI to obtain sulfonic acid, as well as carboxylic acid groups, thereby providing a primarily strong cation exchange with secondary weak cation and weak anion exchange behavior.



HIGH IONIC CAPACITY

Our BAKERBOND® line of polymeric resins were all designed with higher ionic capacities than the industry leading ion exchange and hydrophobic interaction chromatography (HIC) resins. The high ionic capacity increases the salt gradient needed to separate similar molecules, thus improving selectivity. This also enables operations at high salt concentrations. A comparison of ionic capacity between BAKERBOND® PolyCSX™ and several other leading competitive strong cation exchange resins can be seen below.



EASE OF USE

Our BAKERBOND® line of polymeric resins are based on a highly cross linked rigid spherical polymethacrylate media that provides optimal porosity, high mechanical strength, and excellent chemical resistivity delivered with a consistent narrow particle size distribution. These features enable robust resin application, narrow elution bands for concentrated product fractions, and provide consistency and stability to downstream operations.

As illustrated in Fig.2 below, BAKERBOND® PolyCSX™ has an average pore size of 500 Å for an exclusion limit of 1×10^6 dalton. This open porosity coupled with the bead's mechanical strength allows for the use of PolyCSX™ at high linear velocities due to good mass transfer.

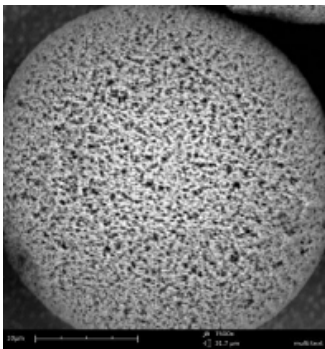
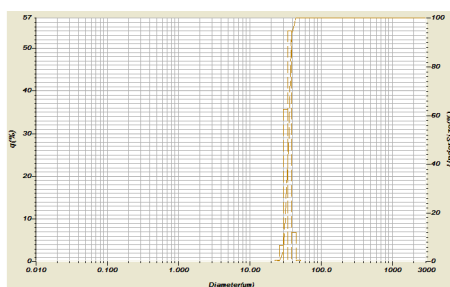


Fig.2. SEM image of polymethacrylate spherical beads at 2,000 magnification

The average pore size of the beads is 500 Å, providing an exclusion limit of 1×10^6 dalton

A small average particle size of 35 µm combined with a narrow particle size distribution (d_{50}/d_{10}) produces narrow elution bands of highly concentrated product for improved efficiency. This increases resolution and decreases pool volumes compared to media based on larger particles. High efficiency in combination with high selectivity enables higher loading levels while maintaining separation.

Fig.3. Particle size distribution of a typical lot of polymethacrylate beads



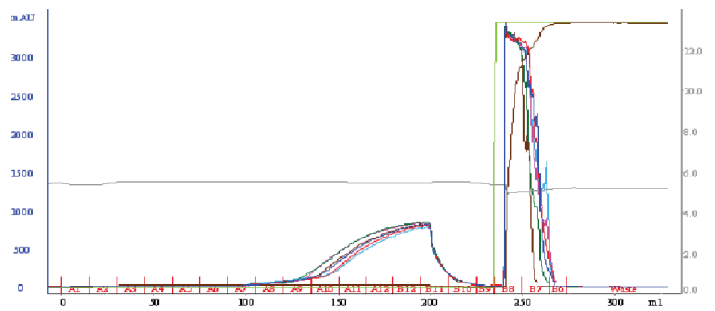
A median particle size of 35 micron and a particle size distribution of less than 1.4 (d_{50}/d_{10}) show a lack of fines and result in high efficiency at low back pressure.

The hydrophilic backbone of BAKERBOND® PolyCSX™ has low non-specific binding and the ability to withstand prolonged contact with commonly used cleaning and sanitizing solutions. PolyCSX™ media can be easily packed to bed heights of up to 40 cm and operated in the linear velocity range of up to 500 cm/hr using conventional columns with pressure ratings of 2 to 10 bar. Our entire line of BAKERBOND® chromatography resins, buffers, and additive are manufactured under the strictest controls and are supported by world class quality systems and application resources.

STABILITY AND CONSISTENCY

The performance of BAKERBOND® PolyCSX™ remains consistent across a broad range of storage conditions. As illustrated in Fig.4, little to no change was identified in protein binding capacity after prolonged storage across a wide range of pH's and temperatures. Product performance was evaluated by measuring the dynamic binding capacity of rabbit IgG. Fig.5 illustrates the purification behavior of PolyCSX™ showed little to no change in its ability to separate a mixture of ovalbumin, rabbit gamma globulin and Lysozyme after subjecting the column to a comparable range of storage conditions.

Fig.4. PolyCSX™ maintains capacity of rabbit IgG in a wide pH range



Storage conditions at extreme pHs have little effect on dynamic binding capacity.

Sample: ovalbumin, rabbit gamma globulin and lysozyme.

Column: 10 . 100 mm packed with PolyCSX™.

Binding buffer: 137 mM HAC/TEA 32.9, mM NaCl pH 4.7

Elution buffer: 1.0 M NaCl in Bind.

Buffer, Gradient: 0 to 100% elution buffer in 10 CV

Flow rate: 5 mL/min.

Blue: 50 days in storage solution at 7 °C

Red: 50 days in storage solution at 47 °C

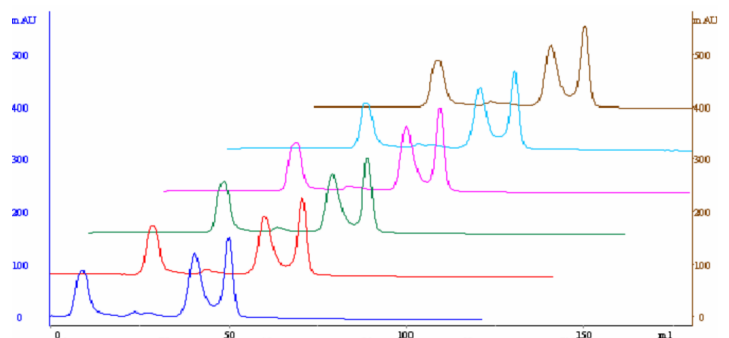
Green: 50 days in 50 mM phosphoric acid at 47 °C

Pink: 50 days in 0.1 M NaOH at 47 °C.

Cyan: 48 hours of washing with 50 mM phosphoric acid at 20 °C

Brown: 48 hours of washing with 0.1 M NaOH at 20 °C.

Fig.5. Chemical stability over a wide pH range



Resolution is not affected by extreme pH conditions over extended periods of time.

OPERATIONAL FLEXIBILITY

BAKERBOND® PolyCSX™ scales easily to the production environment with reproducible results in columns ranging from 20 to 45 cm in internal diameter. The narrow particle size distribution and mechanical stability of the media maintains conventional low pressure column performance at bed heights up to 20 cm. Modern medium pressure columns, rated at 6 bar, can be used to pack beds up to 40 cm in height and enable operation at high linear velocities.

The linear pressure-flow curve for BAKERBOND® PolyCSX™ indicates that there is no resin compression at a bed height of 21.5 cm and a column diameter of 20 cm. In addition, the media does not need to be defined prior to the initial packing or subsequent packings, reducing the consumption of buffers and simplifying operations.

Fig.6. Pressure-Flow relation

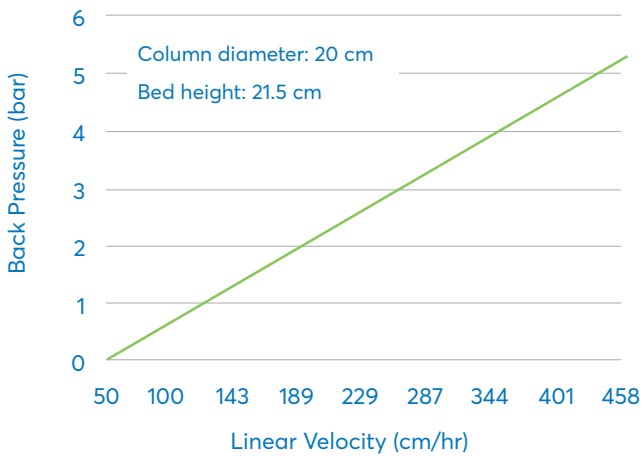


Fig.7. Efficiency of column (20 id x 21.5 cm high) packed with PolyCSX

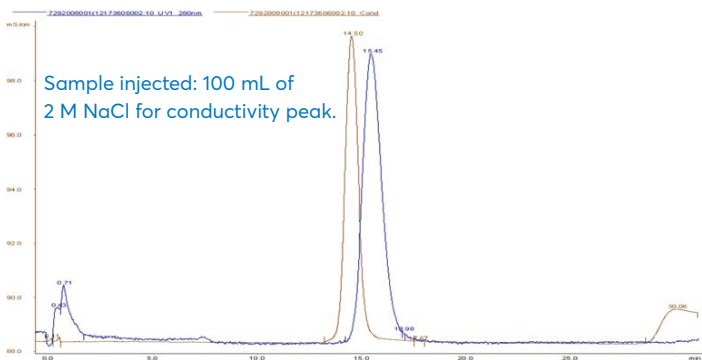


Fig.8. Polymeric media packed in a 200/500 Glass Column



CONVENIENT PACKAGING

BAKERBOND® PolyCSX™ resin comes conveniently packaged in a non-hazardous, non-flammable storage buffer, eliminating many of the burdensome shipping, handling, and storage requirements common among chromatography resins.

Product ordering information

Size	Product Number
1 mL Lab Columns (5-pack)	6061-07
5 mL Lab Columns (5-pack)	6061-25
50 mL	7587-01
100 mL	7587-02
500 mL	7587-03
1 L	7587-04
5 L	7587-05

PROCESS AND APPLICATION SUPPORT

Avantor has deep expertise in process chromatography optimization and can work with you to help ensure that BAKERBOND® PolyCSX™ delivers the improved performance you need in downstream processing. Technical support from our scientists and application specialists is available from our multiple global research and innovation centers.

Product Information Summary

Characteristics

Functionality	Primarily a strong cation exchanger with weak cation and weak anion exchange	
Functional Group	-NH-C(=O)-CH ₂ -CH(SO ₃ H)(CO ₂ H) and equivalent variations	
Ion Exchange Capacity	Cation exchange: 0.3–0.5 H+meq/mL	Anion exchange: 0.10 – 0.20 Cl
Median Particle Size	35 µm	
Particle Size Distribution (d ₆₀ /d ₁₀)	< 1.4	
Median Pore Size	500 Å	
Exclusion Limit	1 × 10 ⁶ Daltons	
Operating pH Range	4.5 – 14.0	
Cleaning pH Range	1.0 – 14.0	
Chemical Stability	All commonly used aqueous buffers, sodium hydroxide, acetic acid, phosphoric acid, guanidine hydrochloride, up to 100% ethanol, methanol, or 2-Propanol.	
Shipping Solvent	Media is shipped as 1:1 slurry in acetate buffer containing benzyl alcohol at pH 4.5 and can be stored for up to 5 years at 4–15 °C. The media can be stored in shipping buffer or 0.1 M NaOH for up to 5 years.	



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