02

Ion Exchange Columns and Media

Ion exchange columns

BioPro IEX Columns

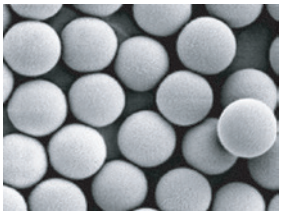
- Hydrophilic polymer beads with low nonspecific adsorption
- Ultra-fast analysis and high resolution analysis on non-porous type
- Ideal for analysis and laboratory scale purification on porous type with high binding capacity and high recovery of biomolecules
- Suitable for characterization of biopharmaceuticals and quality control

■ Usable pH range : 2-12

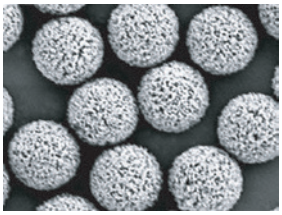
Ion exchange columns for separation of proteins, peptides, and nucleic acids

BioPro IEX columns are specially designed for separation of proteins, peptides, and nucleic acids.
BioPro IEX QF/SF are based on non-porous hydrophilic polymer beads with high chemical and mechanical stability, and low nonspecific adsorption of biomolecules. The short columns (30 mm, 50 mm) are useful for the fast analysis at a higher flow rate, and the 100 mm length columns are best choice for the quality control assessment of biopharmaceuticals requiring a high resolution.
BioPro IEX QA/SP are based on porous hydrophilic polymer beads. BioPro IEX QA/SP have superior resolution, high binding capacity and high recovery of various biomolecules, and they allow highly effective analysis and laboratory scale purification of biopharmaceutical proteins such as antibodies.

SEM images of polymer beads



Non-porous polymer beads



Porous polymer beads

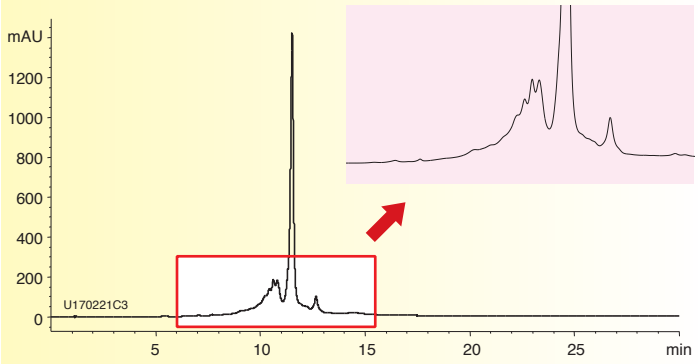
Specifications

	BioPro IEX QF	BioPro IEX SF	BioPro IEX QA	BioPro IEX SP
Matrix	Hydrophilic non-porous polymer		Hydrophilic porous polymer	
Particle size (μm)	3, 5		5	
Charged group	$-CH_2N^+(CH_3)_3$	$-CH_2CH_2CH_2SO_3^-$	$-CH_2N^+(CH_3)_3$	$-CH_2CH_2CH_2SO_3^-$
Counter ion	Cl^-	Na^+	Cl^-	Na^+
Ion exchange capacity* (meq/mL-resin)	0.09	0.24	0.09	0.09
Dynamic binding capacity* (mg/mL-resin)	>12 (BSA)	>10 (human-IgG)	>110 (BSA)	>70 (human-IgG)
Usable temperature	4-60℃			
Usable pH range	2-12			
Column material	PEEK			

*Reference value

High resolution analysis of monoclonal antibody (MAb) <1>

BioPro IEX SF 3 µm, 100 X 4.6 mmI.D.

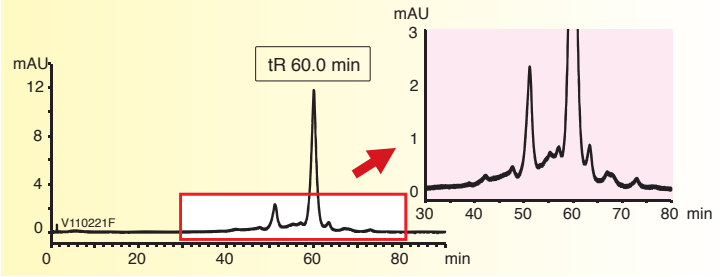


Eluent	: A) 20 mM NaH_2PO_4-Na_2HPO_4 (pH 6.8) B) 20 mM NaH_2PO_4-Na_2HPO_4 (pH 6.8) containing 0.2 M NaCl 0-50%B (0-30 min)
Flow rate	: 0.5 mL/min (180 cm/hr)
Temperature	: 25°C
Detection	: UV at 215 nm
Injection	: 10 µL
Sample	: Humanized monoclonal IgG1 (2.5 mg/mL)

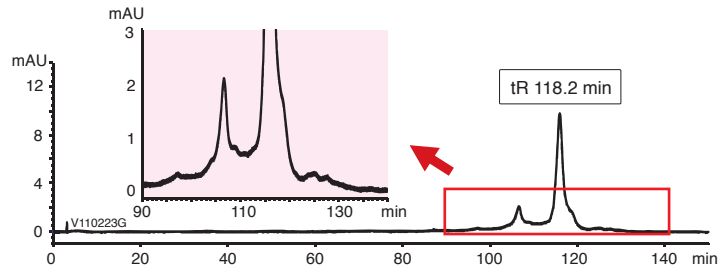
BioPro IEX SF column with 3 µm, 100 mm length achieved high resolution analysis of MAb.

High resolution analysis of monoclonal antibody (MAb) <2>

BioPro IEX SF 5 µm, 100 X 4.6 mmI.D.



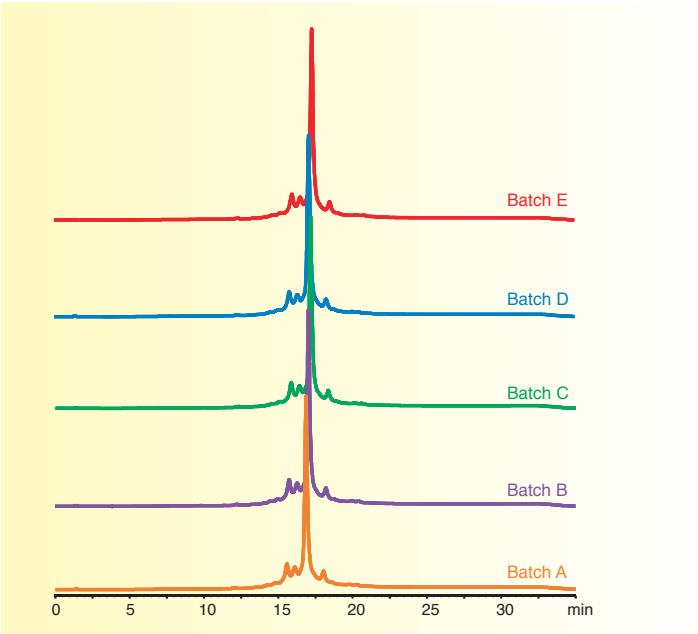
Competitor's WCX column 10 µm, 250 X 4.0 mmI.D.



Eluent	: A) 20 mM MES-NaOH (pH 5.6) B) 20 mM MES-NaOH (pH 5.6) containing 0.2 M NaCl
Initial gradient conc.	: 35%B (70 mM NaCl)
Gradient slope	: 0.25%B/min (0.5 mM NaCl)
Flow rate	: 180 cm/hr (0.5 mL/min for 100 X 4.6 mmI.D., 0.378 mL/min for 250 X 4.0 mmI.D.)
Temperature	: 30°C
Detection	: UV at 280 nm
Injection	: 10 µL
Sample	: Humanized monoclonal IgG1 (1 mg/mL)

The separation of MAb is compared on BioPro IEX SF and competitor's column under the same gradient conditions at pH 5.6. BioPro IEX SF column provides higher resolution of MAb in a shorter analysis time than the competitor's column.

Excellent batch-to-batch reproducibility

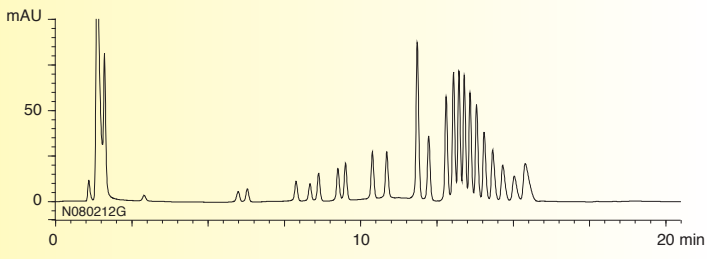


Column	: BioPro IEX SF 5 µm, 100 X 4.6 mmI.D.
Eluent	: A) 20 mM NaH_2PO_4-Na_2HPO_4 (pH 6.5) B) 20 mM NaH_2PO_4-Na_2HPO_4 (pH 6.5) containing 0.2 M NaCl 0-50%B (0.5-30 min)
Flow rate	: 0.5 mL/min (180 cm/hr)
Temperature	: 25°C
Detection	: UV at 215 nm
Injection	: 10 µL
Sample	: Humanized monoclonal IgG1

BioPro IEX SF column exhibits excellent batch-to-batch reproducibility for MAb analysis, including the resolution of small peaks for charge variants. All the medium batches are inspected by various quality control tests and must pass rigorous criteria before release. BioPro IEX columns are the best choice for the quality control of MAbs and other biopharmaceuticals.

High resolution analysis of nucleic acids

BioPro IEX QF 5 µm, 100 X 4.6 mmI.D.



DNA fragments 1Kb DNA ladder (75-12,216 bp)

Eluent	: A) 20 mM Tris-HCl (pH 8.1) containing 0.7 M NaCl B) 20 mM Tris-HCl (pH 8.1) containing 1.0 M NaCl 0-100%B (0-30 min)
Flow rate	: 0.5 mL/min (180 cm/hr)
Temperature	: 25°C
Detection	: UV at 260 nm
Injection	: 20 µL (0.25 mg/mL)

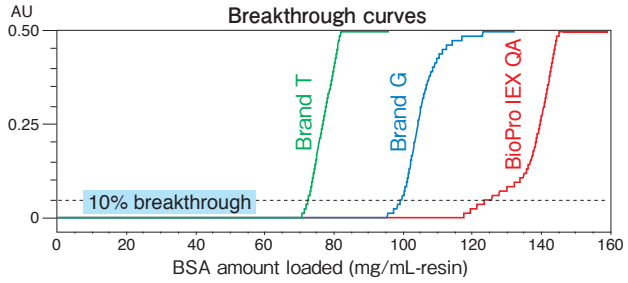
The separation of DNA fragments is shown. BioPro IEX QF of 100 mm length column is a good choice for high-resolution analysis of nucleic acids.

High binding capacity and recovery

Comparison of dynamic binding capacity (DBC) and recovery for BSA

	DBC (mg/mL-resin, 10% breakthrough)	Eluted amount (mg/mL-resin)	Recovery* (%)
BioPro IEX QA	126	120	95
Brand T (porous Q type)	73	58	79
Brand G (porous Q type)	100	35	35

*Recovery: (Eluted amount/DBC) X 100



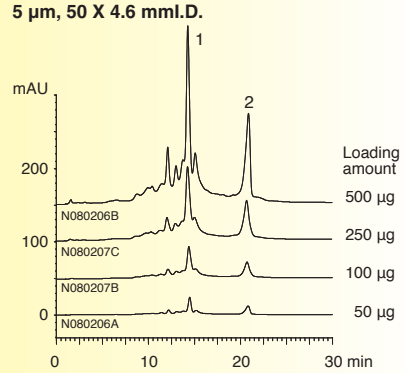
Column	: BioPro IEX QA, 50 X 4.6 mmI.D. Brand T (porous Q type) 50 X 4.6 mmI.D. Brand G (porous Q type) 50 X 5.0 mmI.D.
Linear velocity	: 180 cm/hr
Equilibration buffer	: 20 mM Tris-HCl (pH 8.6)
Elution buffer	: 20 mM Tris-HCl (pH 8.6) containing 1.0 M NaCl
Detection	: UV at 280 nm
Sample	: 1 mg/mL Bovine serum albumin (BSA) in equilibration buffer

BioPro IEX QA gives the superior DBC and recovery compared with conventional porous polymer anion exchange columns. The surface structure of porous type BioPro IEX, which is designed for maximum interaction with proteins, provides high binding capacity, and the hydrophilic property of polymer beads significantly reduces nonspecific adsorption of proteins.

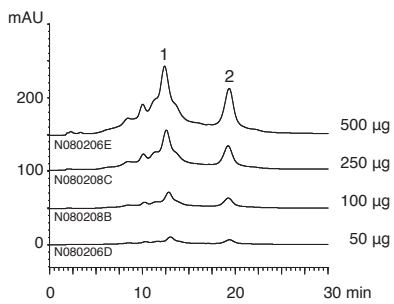
High loadability

Comparison of the effect of sample load on BioPro IEX QA and commercial Q type column

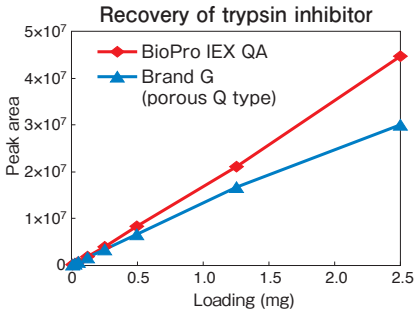
BioPro IEX QA
5 µm, 50 X 4.6 mmI.D.



Brand G (porous Q type)
10 µm, 50 X 5.0 mmI.D.



1. Ovalbumin
2. Trypsin inhibitor



Eluent	: A) 20 mM Tris-HCl (pH 8.1) B) 20 mM Tris-HCl (pH 8.1) containing 0.5 M NaCl 10-80%B (0-30 min)
Flow rate	: 0.5 mL/min (180 cm/hr for 4.6 mmI.D., 150 cm/hr for 5.0 mmI.D.)
Temperature	: 25°C
Detection	: UV at 280 nm
Injection	: 100 µL

BioPro IEX QA shows excellent resolution and peak shapes even when the loading amount increases. The porous type BioPro IEX columns are suitable for laboratory-scale purification of proteins.

Ion exchange media

BioPro IEX SmartSep Q/S
BioPro IEX Q/S

- High productivity on purification
- Hydrophilic polymer beads with low nonspecific adsorption
- High binding capacity and high resolution over a wide range of flow rate
- Suitable for purification of antibodies, proteins and nucleic acids

■ Usable pH range : 2-12

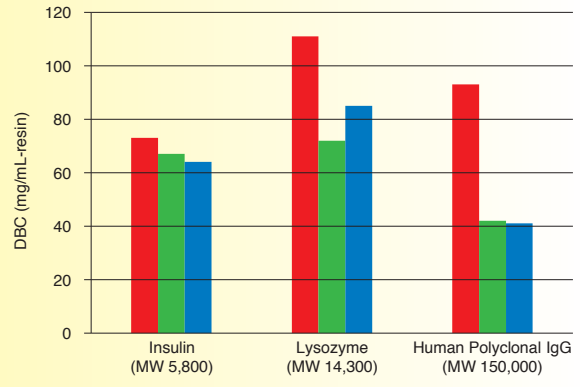
Ion exchange media for purification of biopharmaceuticals

BioPro IEX SmartSep Q/S and BioPro IEX Q/S, strong ion exchange media, are suitable for downstream chromatographic purification processes in biopharmaceutical manufacturing. High dynamic binding capacity and high recovery of those media allow fast purification processes at large scale. It offers high productivity on industrial purification.
BioPro IEX SmartSep Q/S, which show high resolution and recovery even at a high flow rate and high loading condition, are suitable for an intermediate purification step and a polishing step of MAbs, proteins, peptides, and oligonucleotides.
BioPro IEX Q/S are designed for capture and intermediate purification of biopharmaceuticals such as MAbs.

Specifications

	BioPro IEX SmartSep Q	BioPro IEX SmartSep S	BioPro IEX Q	BioPro IEX S
Matrix	Hydrophilic porous polymer			
Particle size (µm)	10, 20, 30		75	
Charged group	$-R\text{-}N^+(CH_3)_3$	$-R\text{-}SO_3^-$	$-R\text{-}N^+(CH_3)_3$	$-R\text{-}SO_3^-$
Ion exchange capacity (meq/mL-resin)	>0.08		>0.10	
Dynamic binding capacity (mg/mL-resin)	>100 (BSA)	>100 (lysozyme)	>160 (BSA)	>160 (lysozyme)
Usable pH range	2-12			

High dynamic binding capacity (DBC) for various samples



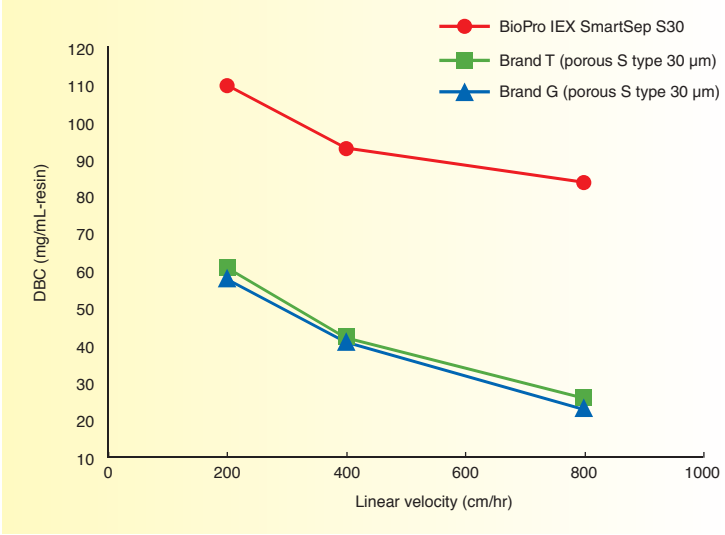
Conditions of DBC measurement*
Column : 50 X 5.0 mmI.D.
Flow rate : 400 cm/hr (1.32 mL/min)
Temperature : 25°C

*Inquire us for details.

	Particle size (µm)	DBC (mg/mL-resin, 10% breakthrough)		
		Insulin	Lysozyme	Human Polyclonal IgG
BioPro IEX SmartSep S30	30	73	111	93
Brand T (porous S type)	30	67	72	42
Brand G (porous S type)	30	64	85	41

BioPro IEX media have higher DBC compared to conventional ion exchange media. Especially for IgG, BioPro IEX media have more than twice as high DBC as competitors' media. This feature of BioPro IEX media makes purification productivity of IgG per unit time double or more.

DBC over a wide range of flow rate

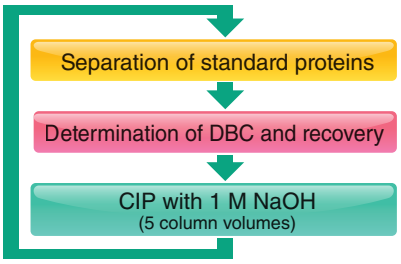


Column	: 50 X 5.0 mmI.D.
Equilibration buffer	: 20 mM citric acid-NaOH (pH 5.3)
Elution buffer	: 0.5 M NaCl in equilibration buffer
Flow rate	: 200-800 cm/hr (0.66-2.62 mL/min)
Temperature	: ambient (25℃)
Detection	: UV at 280 nm
Sample	: 1.5 mg/mL human polyclonal IgG in equilibration buffer

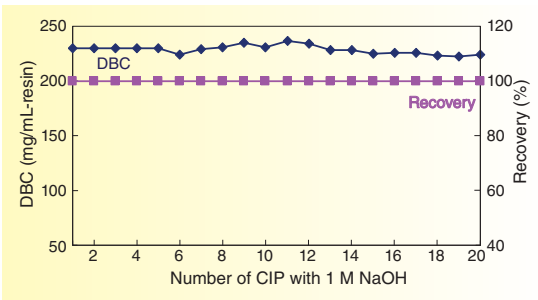
High DBC of BioPro IEX media is maintained even at higher flow rate, making them suitable for the high-speed purification with 2-3 times of conventional flow rates. This feature offers significant improvement on productivity.

Excellent durability (Stability on CIP)

Test protocols



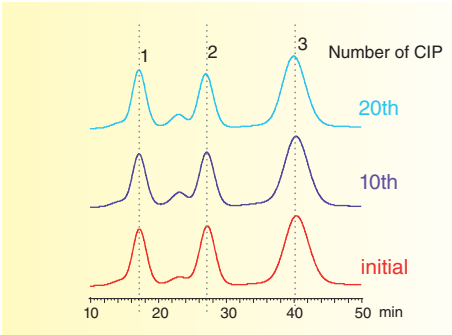
DBC and recovery



Conditions of DBC* measurement	
Column	: BioPro IEX S75, 50 X 5.0 mmI.D.
Flow rate	: 800 cm/hr (2.62 mL/min)
Equilibration buffer	: 20 mM Glycine-NaOH (pH 9.0)
Elution buffer	: 0.5 M NaCl in equilibration buffer
Sample	: 1.0 mg/mL Lysozyme in equilibration buffer
Temperature	: ambient
Detection	: UV at 300 nm

*DBC was determined at 10% breakthrough

Separation of standard proteins

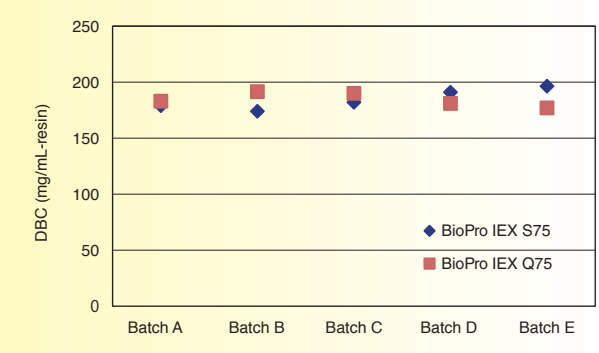


1. Ribonuclease A
2. Cytochrome *c*
3. Lysozyme

Conditions of separation of standard proteins	
Column	: BioPro IEX S75, 50 X 5.0 mmI.D.
Eluent	: A) 20 mM NaH_2PO_4-Na_2HPO_4 (pH 6.8) B) 20 mM NaH_2PO_4-Na_2HPO_4 (pH 6.8) containing 0.5 M NaCl
Gradient	: 0-100%B (0-60 min; Linear)
Flow rate	: 180 cm/hr (0.59 mL/min)
Temperature	: 25°C
Detection	: UV at 220 nm
Injection	: 24 µL (0.5 mg/mL)

Cleaning in place (CIP) is an important procedure for cleaning and sterilization of columns used for protein purification. The DBC and the selectivity of proteins are unaffected following 20 cycles of CIP with 1 M NaOH. The high chemical stability of BioPro IEX media allow effective cleaning with alkaline solution.

Excellent batch-to-batch reproducibility of DBC



Column	: 50 X 4.6 mmI.D.
Flow rate	: 180 cm/hr
for anion-exchange resin	
Equilibration buffer	: 20 mM Tris-HCl (pH 8.6)
Elution buffer	: 0.5 M NaCl in equilibration buffer
Sample	: 1.5 mg/mL BSA in equilibration buffer
Detection	: UV at 280 nm
for cation-exchange resin	
Equilibration buffer	: 20 mM Glycine-NaOH (pH 9.0)
Elution buffer	: 0.5 M NaCl in equilibration buffer
Sample	: 1.5 mg/mL Lysozyme in equilibration buffer
Detection	: UV at 300 nm

BioPro IEX media exhibit excellent batch-to-batch reproducibility of DBC. All the medium batches are inspected by various quality control tests. We supply stable products over a long period of time.

BioPro Ion Exchange Screening Kit

The BioPro Ion Exchange Screening Kit is a set of screening columns packed with BioPro IEX media.

- Two column types (1 mL and 5 mL) ideal for media screening, development of purification methods, and loadability studies
- Easy installation and convenient use

Ordering Information –Columns–

BioPro IEX QF/SF

Phase dimension	Column I.D. (mm)	Column length (mm)		
		30	50	100
BioPro IEX QF non-porous 3 µm	4.6	QF00S03-0346WP	QF00S03-0546WP	QF00S03-1046WP
BioPro IEX QF non-porous 5 µm	4.6	QF00S05-0346WP	QF00S05-0546WP	QF00S05-1046WP
BioPro IEX SF non-porous 3 µm	4.6	SF00S03-0346WP	SF00S03-0546WP	SF00S03-1046WP
BioPro IEX SF non-porous 5 µm	4.6	SF00S05-0346WP	SF00S05-0546WP	SF00S05-1046WP

BioPro IEX QA/SP

Phase dimension	Column I.D. (mm)	Column length (mm)		
		30	50	100
BioPro IEX QA porous 5 µm	4.6	QAA0S05-0346WP	QAA0S05-0546WP	QAA0S05-1046WP
BioPro IEX SP porous 5 µm	4.6	SPA0S05-0346WP	SPA0S05-0546WP	SPA0S05-1046WP

Ordering Information

BioPro IEX media

Product name	Particle size (µm)	Product number
BioPro IEX SmartSep Q10	10	QSA0S10
BioPro IEX SmartSep S10		SSA0S10
BioPro IEX SmartSep Q20	20	QSA0S20
BioPro IEX SmartSep S20		SSA0S20
BioPro IEX SmartSep Q30	30	QSA0S30
BioPro IEX SmartSep S30		SSA0S30
BioPro IEX Q75	75	QAA0S75
BioPro IEX S75		SPA0S75

BioPro Ion Exchange Screening Kit

Packing material	Particle size (µm)	Specification	Column volume (mL)	Product number
BioPro IEX SmartSep Q20	20	5/pack	1	BPQSA0S20-01PK
			5	BPQSA0S20-05PK
BioPro IEX SmartSep S20			1	BPSSA0S20-01PK
			5	BPSSA0S20-05PK
BioPro IEX SmartSep Q30	30	5/pack	1	BPQSA0S30-01PK
			5	BPQSA0S30-05PK
BioPro IEX SmartSep S30			1	BPSSA0S30-01PK
			5	BPSSA0S30-05PK
BioPro IEX Q75	75	5/pack	1	BPQAA0S75-01PK
			5	BPQAA0S75-05PK
BioPro IEX S75			1	BPSPA0S75-01PK
			5	BPSPA0S75-05PK