Care and Use Instructions
Ion Exchange Media
BioPro SmartSep Q / BioPro SmartSep S

1. Introduction

Thank you for purchasing YMC products for ion exchange media. BioPro SmartSep Ion Exchange media are based on high mechanical strength hydrophilic polymer beads with a strong-anion exchanger (quatemary ammonium group) or a strong-cation exchanger (sulfobutyl group), and are most suitable for capture and intermediate purification.

BioPro SmartSep Ion Exchange media, which are manufactured under highly controlled conditions, must pass a series of stringent tests before being accepted for shipment (Please refer to the inspection report). To ensure optimal performance and durability of the media, please follow these instructions.

2. Specification

Item	Strong-anion exchanger		Strong-cation exchanger	
	BioPro SmartSep Q10	**BioPro SmartSep Q30**	**BioPro SmartSep S10**	**BioPro SmartSep S30**
Particle size (µm)	10	30	10	30
Matrix	Hydrophilic polymer beads			
Functional group	$-R-N^+(CH_3)_3$		$-R-SO_3^-$	
pH range	2.0 – 12.0		2.0 – 12.0	
Temp. range (°C)	4 – 40		4 – 40	
Pressure range	Regular use: 3 MPa or less than 3 MPa Upper limit: 4 MPa	Regular use: 2 MPa or less than 2 MPa Upper limit: 3 MPa	Regular use: 3 MPa or less than 3 MPa Upper limit: 4 MPa	Regular use: 2 MPa or less than 2 MPa Upper limit: 3 MPa
Shipping solvent	20% ethanol aqueous solution			
Feature	Highly efficient polishing step from intermediate purification step			

3. Packing instructions

3-1 Resin preparation (Removal of fines)

Recommended slurry solvent : Deionized water or 20% ethanol solution

1. The amount of chromatography medium needed is approximately 1.05 X to 1.15 X column volume. Suspend the settled resin in the shipping container(s) and transfer the slurry in to a vessel whose capacity is >5 times larger than the amount of slurry needed to pack the column. Allow the slurry to settle for a certain time and measure the amount of the resin. Decant the supernatant.
2. Add four times the resin volume of above stated slurry solvent to the vessel.
3. Suspend the solution with a rod or paddle. Do not use a sharp-edged paddle or a magnetic stirrer to avoid fine generation.
4. Allow the resin to settle for 120 minutes.
5. Once resin has settled, decant the supernatant.
6. Repeat 2-5 until the supernatant at the Step 5 becomes clear.

3-2 Packing slurry preparation and column packing

Recommended slurry solvent: A solution with high ionic strength (a component of mobile phase which has the highest ionic strength, 1 M NaCl, 0.5 M Na_2SO_4)

1. Filter the slurry. If 20% ethanol is used at 3-1 Resin preparation, wash the resin on the funnel with deionized water (This process can be skipped if deionized water is used at 3-1).
2. Wash the resin with approximately three times the resin volume of above stated slurry solvent.
3. Add the slurry solvent to adjust the slurry concentration to 30-50% and suspend the resin. Gently pour the suspended slurry into the column. Prevent air coming into the column.
4. Start the pump and pack the column at approximately two times the flow rate of the actual flow rate in use. Maintain the flow rate until the packed bed becomes stable.

NOTE) Also refer to the Instruction manual of the column used. Adjust the packing method in accordance with it, when necessary.

3-3 Column performance evaluation

When the column packing is completed, evaluate the column performance evaluation by injecting a sample to the column, and determine the column theoretical plate number (N/m) and peak asymmetry factor (As). Typical evaluation condition is shown below. If there is a large difference between the value(s) obtained and typical performance listed below, adjust the packing parameter(s) and repeat the packing procedure.

Conditions

Mobile phase:	Low ionic strength buffer
	Strong anion exchanger (Q): 20 mM Tris-HCl buffer (pH 8)
	Strong cation exchanger(S): 20 mM Phosphate buffer (pH 7)
Flow rate:	72 cm/h
Temperature:	ambient (25 °C)
Detection:	UV at 220 nm
Sample:	Formamide (2 µL/mL)
Injection volume:	1% of bed volume

Typical column performance

BioPro SmartSep 10 µm		BioPro SmartSep 30 µm	
Plates (N/m):	≧ 20,000	Plates (N/m):	≧ 8,000
Asymmetry (As):	0.7 - 1.4	Asymmetry (As):	0.7 - 1.4

＊ These values are just guide values. Desired separation could be achieved even if the values obtained are out of the range shown above, depending on your usage and/or application.

4. Equilibration and elution

- Equilibrate with about 5–10 column volumes of initial mobile phase before using a column for chromatographic separations.
- Generally, samples are adsorbed onto the top of the column with 20–50 mM of buffer as the first mobile phase, then eluted with a salt-concentration gradient method (sodium chloride concentration commonly adjusted between 0 to 0.5 M) or pH gradient method. It is recommended to flush the column with buffer containing about 1 M of sodium chloride for each run to remove residual impurities from the column with the final mobile phase .
- Water-soluble organic solvent (maximum of 30%), can be added to the mobile phase. Before adding such solvent, make sure that salt in the buffer will not precipitate. Other additives such as urea (≤ 8 M) or guanidine hydrochloride (≤ 6 M), which are commonly used as protein denaturants, nonionic surface-active agents, cationic surface-active agents (limited to BioPro SmartSep Q), or anionic surface-active agents (limited to BioPro SmartSep S) are useful.
- Avoid solvents containing oxidant for the mobile phase.
- Avoid anionic surface-active agents for BioPro SmartSep Q, cationic surface-active agents for BioPro SmartSep S.

5. Cleaning and regeneration

A change of retention time or peak shape and/or pressure increase might result from the adsorption of fat-soluble substances or precipitated impurities in a sample. In such cases, follow these steps for column cleaning and regeneration. If these procedures will not solve the problem, then we recommend that you use new media.

Batch method:	Soak and agitate the media in washing solution about 3–5 times of the media volume. After leaving it stationary, remove the supernatant fluid by decantation. Repeat the process 2–3 times.
Column method:	Flush the column with washing solution of about 3–5 times the media volume (Disconnecting the column from the detector is recommended). After the process, perform sufficient equilibration with the mobile phase. The state of contamination or type of washing solution (high viscosity, etc.) can cause a pressure increase. In such cases, reduce the flow rate for flushing.

Regarding a washing solution, highly concentrated sodium chloride solution (For example, about 1–2 M concentration) is recommended instead of flushing buffer processing for each run. If performance does not recover, wash with sodium hydroxide (about 0.1–0.5 M) at first, then flush with sodium chloride (about 0.1–0.5 M) and replace with mobile phase.

6. Storage

Store the packed or bulk media in 20% ethanol aqueous solution. Store the bulk media in the original container at a temperature of 4 – 35°C. Keep the container closed tightly.

YMC CO., LTD.

YMC Karasuma-Gojo Bldg., 284 Daigo-cho, Karasuma Nishiiru Gojo-dori, Shimogyo-ku, Kyoto 600-8106, Japan
TEL +81-75-342-4515 FAX +81-75-342-4550
URL:www.ymc.co.jp E-Mail:sales@ymc.co.jp