Tinzyme Co., Limited

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PG 200 resin 400ml

Product Number: HN120210100M

Shipping and Storage

4-30℃ in 20% ethanol

Description

PG 200 resin 400ml is a gel filtration chromatography (also called as size exclusion chromatography) media, and is applicable to the fine purification of biomolecules and buffer exchange at laboratory scale and industrial scale.

Matrix	Highly cross-linked sepharose and glucan
Range of particle size	25-45μm
Mean particle size D50	35±5μm
Separation range	10 - 600 kDa (globulin)
pH stability	3-12 (long-term) 1-14 (short-term)
Chemical stability	All common buffers: 8M urea, 6M guanidine hydrochloride, 70% ethanol, 1M sodium hydroxide

Specification

Product Name	Specification	Product Number
PG 200 resin 400ml	25ml	HN120210025M
PG 200 resin 400ml	100ml	HN120210100M
PG 200 resin 400ml	500ml	HN120210500M
PG 200 resin 400ml	1L	HN120210001L
PG 200 resin 400ml	5L	HN120210005L
PG 200 resin 400ml	20L	HN120210020L

Features

1. High pressure resistance and high flow rate.
2. Small particle size and high resolution.
3. Good compatibility, capable of withstanding more severe cleaning conditions.

Preparation of Solution

Eluent: Prepare according to the customer's requirements.

Note: It is recommended to add salt at a certain concentration (at least 0.025M) to the target solution to suppress the ionic interaction between the sample and the media.

Preparation of Sample

The samples are filtered (filter membrane selection: 0.22μm if mean particle size is <45μm; 0.45μm if 45μm < mean particle size < 165μm; 0.8μm if mean particle size is > 165μm).

Purification Process (taking a chromatographic column packed with XK 16/70 as an example)

1. Washing
   1.1. Take 140 g of sedimentation media, resuspend with purified water of 3 times the volume,and then drain with a G3 sand core funnel; repeat this step for twice.
2. Homogenizing
   2.1. Resuspend the washed and drained media with the same volume (≈ 140 mL) of purified water, and slowly stir with a

glass rod to mix well.

3. Preparation of chromatographic column
   - 3.1. Wash the column tube, lower column head, adapter and column packer with purified water;
   - 3.2. Tighten the sealing ring after installing the lower column head;
   - 3.3. Inject purified water to the height of 3 - 5 cm;
   - 3.4. Unscrew the lower plug to exhaust all air bubbles, then screw the lower plug;
   - 3.5. Install the column packer, and vertically fix the column near the chromatography system.
4. Degassing
   - 4.1. Degas the homogenized media with a vacuum pump or ultrasonic cleaner.
5. Column media
   - 5.1. After stirring the degassed media gently with a glass rod or other stirrer apparatus,add the media (drained with the glass rod to avoid introducing air bubbles) into the chromatographic column quickly and continuously.
   - 5.2. Quickly fill the upper end of the column packer with purified water, and install the packer cap.
   - 5.3. After connecting the chromatography system to the column packer with a tube lines,unscrew the lower plug, press the column at a flow rate of 3 mL/min (90 cm/h) for 120min, and then suspend the chromatography system.
   - 5.4. Screw the lower plugs, quickly remove the column packer, and fill the column tube with purified water.
   - 5.5. Connect the adapter to the chromatography system; after the air bubbles in the tube lines are exhausted, tilt the adapter slightly and insert it into the column tube to the position 0.5 cm - 1 cm away from the upper end of the media interface, then tighten the sealing ring.
   - 5.6. Unscrew the lower plug, press the column at a flow rate of 7 mL/min (210 cm/h) for 30min, and then stop the chromatography system.
   - 5.7. Mark the media interface, and press the adapter to 0.3 cm below the media interface.
6. Column efficiency test
   - 6.1. Connect the chromatographic column outlet to the chromatography system, and equilibrate 2 CV (column volume) with purified water at a flow rate of 1 mL/min (30cm/h);
   - 6.2. Inject 1% CV of 2% acetone with a quantitative loop or injection pump;
   - 6.3. Run 1.5 CV at a flow rate of 1 mL/min (30 cm/h);
   - 6.4. Calculate HETP and As; the column efficiency result is qualified if HETP $\leq$ 3 h (h is the mean particle size) and 0.7 $\leq$ As $\leq$ 1.3, otherwise the column shall be reinstalled.
7. Use
   - 7.1. The column can be used immediately if the column efficiency result is qualified; equilibrate 2 CV with equilibration solution (add 0.15M sodium chloride to the equilibration solution to inhibit possible non-specific adsorption) at a flow rate of 1 mL/min (30 cm/min), and then inject the sample (the injection volume is recommended to be $\leq$ 2.5%CV) for separation; if the column efficiency result is qualified, equilibrate 2 CV with the preservation solution at a flow rate of 1 mL/min (30 cm/min), and store the column.

Washing (taking a chromatographic column packed with XK 16/70 as an example)

1. Wash 2 CV with purified water at 1 mL/min (30 cm/h).
2. Wash 2 CV with 0.5M NaOH at a flow rate of 1 mL/min (30 cm/h).
3. Wash 2 CV with purified water at a flow rate of 1 mL/min (30 cm/h)
4. Wash 2 CV with 0.5M acetic acid at a flow rate of 1 mL/min (30 cm/h).
5. Wash 2 CV with purified water at a flow rate of 1 mL/min (30 cm/h)
6. Wash 2 CV with 30% isopropanol at a flow rate of 1 mL/min (30 cm/h).
7. Wash 2 CV with purified water at a flow rate of 1 mL/min (30 cm/h)
8. Wash 2 CV with 20% ethanol at a flow rate of 1 mL/min (30 cm/h).

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Disinfection

1. Wash 2 CV with purified water at 1 mL/min (30 cm/h).
2. Back-rinse 2 CV with 0.5M NaOH at a flow rate of 1 mL/min (30 cm/h).
3. Wash 2 CV with purified water at a flow rate of 1 mL/min (30 cm/h)
4. Wash 2 CV with 20% ethanol at a flow rate of 1 mL/min (30 cm/h).

Common Problems

Common problems and solutions

Problem	Possible causes	Solution
Poor resolution between the target peak and the impurity peak	1. The injection volume is too large	Decrease the injection volume to 0.5%CV
	2. The sample has high viscosity	Dilute the sample appropriately
	3. The flow rate is too high	Decrease the flow rate
	4. The chromatographic column is too short	Select a longer or thinner chromatographic column
	5. The dead volume is too large	Minimize the dead volumes in at tube lines and fittings
	6. Poor column media	Re-pack the column, or use a pre-packed column
	7. The sample is not filtered	Filter the sample with 0.22 μm or 0.45μm filter membrane
	8. The media is too dirty	Clean and re-equilibrate the column
	9. The column is not installed vertically	Re-pack the column
	10. Uneven temperature during use	It is recommended to maintain a constant temperature
No expected elution peak	1. The injection volume is not the same as before	Maintain the same injection volume
	2. There is an ion interaction between the protein and the media	Maintain the ionic strength of the buffer at 0.05 - 0.15 NaCl
	3. There is a hydrophobic interaction between the protein and the media	The hydrophobic interaction can be minimized by lowering the ionic strength or by increasing the pH, and adding detergents or organic reagents
	4. There is any change in the sample during storage	Prepare the sample freshly before use
	5. Proteins and lipids precipitate in the column	Wash the column or replace with a new column
	6. Microbial growth in the media	The column will be free from microbial growth during use, and must be stored with 20% ethanol.
Elution peak appears earlier	1. There are gaps in the packed column	Re-pack the column
	2. Proteins form dimers or multimers	Pay attention to maintain sample stability under experimental conditions
Elution peak is delayed	1. There is an ion interaction or hydrophobic interaction between the protein and the media.	Maintain the ionic strength of the buffer at 0.05 - 0.15 NaCl

	2. Dirty media, filter membrane, or the top of column bed	Clean and re-equilibrate the column
	3. Microbial growth in the media	The column will be free from microbial growth during use, and must be stored with 20% ethanol.
Chromatographic peaks rise slowly	The media is packed at higher concentration	Re-pack the column
Chromatographic peaks rise slowly or with tailing	The media is packed at lower concentration	Re-pack the column
Cracked or dry column bed	There is any leakage, or large air bubbles are introduced	Check the tube lines for leakage or air bubbles, and re-pack the column
Slow flow rate	1. Aggregation of proteins or lipids	Clean the media or filter membrane in time
	2. Proteins precipitate in the media	Modify the components of the eluent to maintain the stability of target substance
	3. Microbial growth in the media	The column will be free from microbial growth during use, and must be stored with 20% ethanol.
	4. The column bed is compressed	Re-pack the column
Air bubbles are introduced in the column bed	There is a temperature difference during use, or there is residue in the tube lines	Re-pack the column
Pressure increased	1. The sample is turbid	Prepare the sample freshly before use
	2. The tube lines and sieve plate are blocked	Clean the tube lines, sieve plate and media, and re-pack the column