

Operating Instructions

Strong Anion Exchange Chromatography Media Cellufine Q-500

Description

Cellufine Q-500 media is designed for the anion exchange chromatography of acidic proteins, peptides and other biomolecules. The resins are comprised of beaded spherical cellulose, functionalized with a quarternary amine (trimethy-laminoethyl).

The pore size and structure of each media determines its respective applications. Cellufine Q-500 medium is ideal for molecules up to 500 kD. The improved rigidity of Cellufine allows for high flow rates, and thus, rapid processing times, even in large diameter process scale columns.

Physical-Chemical Characteristics

Support matrix	cellulose
Particle shape	spherical
Particle diameter (µm)	ca.40 – 130
Ion capacity (meq/g dry)	1.5
BSA capacity (mg/ml)	> 10
MW exclusion limit (kD)	500
pH stability range	2 - 12
Operating pressure	< 2 bar (29 psi)
Supplied	suspension in 20 % EtOH

Column Packing

1. Calculate volume required of the desired bed dimension.
2. Prepare a 40 – 60 % (v/v) slurry with water, 0.1 M NaCl solution and the appropriate elution buffer. Allow to equilibrate at ambient temperature for one hour.
3. Gently stir or place under vacuum to degas.
4. With column outlet closed, carefully pour the slurry into the column. Depending on the volume, a filler tube may be necessary.
5. With the inlet open to release air, insert and affix the top adjuster. Assemble at the slurry interface.
6. Open the column outlet and begin pumping elution buffer at rate 10 % – 20 % greater than the operational flow rate.

7. After the bed stabilizes, close the column outlet. Then with the inlet open, reposition the end cell on top of the bed. Equilibrate with 10 column volumes of adsorption buffer before sample loading.

Operating Guidelines

General Operation

Typically, adsorption to Cellufine Anion Exchange medium occurs in relatively low ionic strength (e.g., < 0.1 M NaCl) in the pH range from 6.5 – 8.5. Under these conditions, negatively charged proteins will bind. Bound components are then resolved via either stepwise or linear gradient elution.

Recommended Buffers

Adsorption buffer: 0.02 M sodium phosphate or Tris-HCl (pH 8.0).

Elution buffer: 0.1 – 0.2 M sodium chloride in adsorption buffer.

(Other common buffer systems may be used.)

For additional information on protein purification, see References 1 and 2.

Sample Preparation and Load

Prepare samples at a concentration of 1 – 20 mg/ml, in adsorption buffer. Remove insoluble material by centrifugation or microfiltration. If necessary, exchange sample buffer using dialysis, diafiltration or desalting chromatography.

Recommended Flow Rate: 50 – 200 cm/h.

Chemical and Physical Stability

Stable in:

Most salts (NaCl, (NH₄)₂SO₄, etc.)

Most detergents (SDS, Tween®, Chaps, etc.)

0.5 M NaOH

Autoclavable: 121°C, 20 minutes

Regeneration and Depyrogenation

To regenerate a column, flush bed with 2 - 5 column volumes of 0.5 M NaOH, followed by several volumes of elution buffer. Then equilibrate as usual. If depyrogenation is required, wash the column with 2 - 5 column volumes of 0.5 M NaOH followed by several column volumes of pyrogen free elution buffer. Monitor the pyrogen levels in the column eluate

during a blank gradient elution prior to reusing the column.

Storage

Store unopened container at ambient temperature. Do not freeze.

Short term storage for bulk and column (2 weeks or less) can be at a room temperature with 0.05 M NaOH. Longer storage should be in neutral buffer containing 0.02 % sodium azide or 20 % ethanol, at 2 – 8 °C. Do not freeze.

Shelf Lifetime:

5 years from date of manufacture

References

1. Janson, J. C. and Ryden, L., *Protein Purification: Principles, High Resolution Methods and Applications*. VCR Publications Inc., 23 rd Street, NY (1989)
2. Harris, E.L.V. and Anfalg, S., *Protein Purification Methods: A practical Approach*. New York: Oxford University Press, New York (1989).

Product Ordering Information (Catalogue No.)

Media type	Pack Size				
	Mini-Column 1ml x 5	100ml	500ml	5 lt	10 lt
Cellufine Q-500	19907-51	675 982 327	19907	19908	675 982 335
Cellufine A-200		676 980 327	19611	19612	676 980 335
Cellufine A-500	19805-51	675 980 327	19805	19806	675 980 335
Cellufine A-800		673 980 327	19800	19801	673 980 335
Cellufine C-500	19800-51	675 983 327	19865	19866	675 983 335

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