

## Operating Instructions



IN008EN Ver1.3

**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 quaternary amine (trimethylaminoethyl).

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 ( $\mu\text{m}$ ) | 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 the appropriate elution buffer (high salt). 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,  $(\text{NH}_4)_2\text{SO}_4$ , 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

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 Anfgal, 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<br>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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