

Ion Exchange Chromatography Media

Cellufine IEX

Technical Data Sheet



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Technical DATA Sheet

Cellufine Ion Exchange Media

What is Cellufine?

Cellufine is porous and spherical chromatography medium manufactured from cellulose. Cellufine has been approved in many bio-pharmaceutical GMP production processes since 1981. Cellulose is well-known as natural product having unique crystalline molecular structure which brings to have chemicals stability and mechanical strength. Owing to these properties, Cellufine has an excellent column packing characteristics and can be packed at high flow rates due to high mechanical strength of produced particles.

Cellufine IEX Media

Cellufine is available with six different IEX media as shown in Table 1 and their ligand structure are described in Fig. 1. Cellufine IEX 500 series media in Table 1 is used as conventional typed media.

The pore property (pore size) of media influences on the performance of chromatography medium. Fig. 2 shows the relationship Kav profile on each base resin of Cellufine IEX media. JNC offers 3 different weak anion media having different from pore property described in the next.

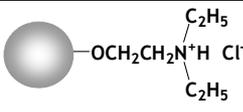
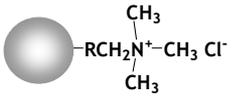
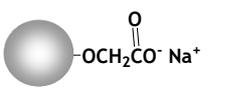
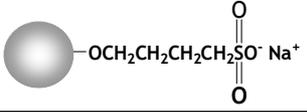
| | |
|----------------------------------|-------------------------------------------------------------------------------------|
| Cellufine A-200, A-500, A-800 |  |
| Cellufine Q-500 |  |
| Cellufine C-500 |  |
| Cellufine S-500 |  |

Fig. 1. Ligand structure of Cellufine IEX media

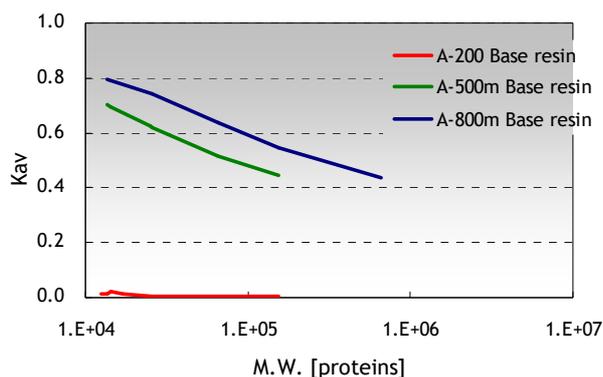


Fig. 2. Kav profile on each base resin of Cellufine anion exchange chromatography

Characteristics of Cellufine IEX Media

The basic characteristics of Cellufine IEX media are shown in Table 1. All Cellufine IEX media are based on 90 μm (average) cross-linked cellulose beads. Figure 3 shows particle-size distributions of Cellufine A-500 which is a standard media as Cellufine IEX 500 series.

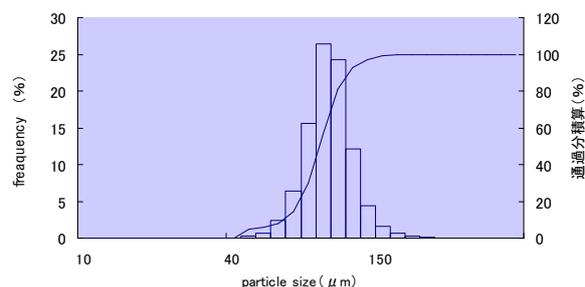


Fig. 3. Particle-size distributions of Cellufine A-500

Cellufine 500 type (Cellufine A-500, Cellufine Q-500, Cellufine C-500 and Cellufine S-500) is a standard medium in Cellufine IEX medium. The media have good flow property for industrial. These are resins with relatively large pore size which is enough effective for IgG.

Cellufine A-200 has the smallest pore size in Cellufine IEC media. As the adsorption and desorption of protein is occurred on the surface of A-200 resins, a typical breakthrough curve in the media is very sharp as shown in Fig. 4. Thus Cellufine A-200 is used as flow through mode with taking advantage of its unique characteristic in adsorption.

Cellufine A-800 has large pore size which makes huge protein such as thyroglobulin (MW=660kDa) stay inside. While Cellufine A-800 media is poor flow property as in Fig. 5, dynamic binding capacity of the medium is

excellent as in Fig. 6. Especially the medium is suitable for the purification of large protein.

Cellufine Q-500 is manufactured from A-500 by attaching tri-methyl ammonium ligand. One of characteristics in the medium is to be able to use under high salt condition as in Fig.9.

Pressure-flow Properties of Cellufine IEX Media

Figure 5 shows pressure-flow velocity curves of Cellufine IEX media in a 2.2 cm column with a 20 cm bed height. Figure 6 also shows pressure-flow velocity curves of Cellufine A-500 media in a 30 cm column with a 20 cm bed height. Cellufine IEX media enable high-flow operation, which is essential to efficient purification of bio-pharmaceuticals.

Table 1. Characteristics of Cellufine IEX media

| | A-200 | A-500 | A-800 | Q-500 | C-500 | S-500 |
|---------------------------|------------------------|-----------|-----------|-------------|-----------|------------|
| Base Matrix | Cross-linked cellulose | | | | | |
| Particle size (μ m) | 40-130 | | | | | |
| Ion exchange type | Weak (DEAE) | | | Strong (QA) | Weak (CM) | Strong (S) |
| Ion Exchange (meq/ml-gel) | 0.13-0.18 | 0.13-0.17 | 0.05-0.08 | 0.14-0.29 | 0.09-0.12 | 0.11-0.22 |

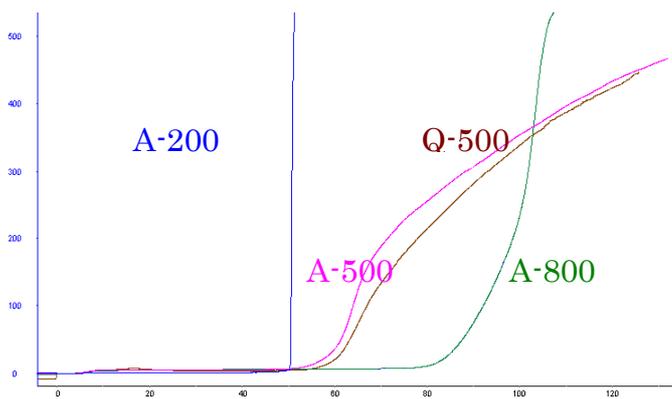


Fig.4. Typical break through curves of BSA for Cellufine anion IEX media

Column: 5 mm ID \times 50 mm L

Flow rate: 150 cm/h

Sample: 1 mg/ ml

Buffer: 50 mM Tris-HCl (pH 8.5) for A-200, A500 and A-800

50 mM Tris-HCl (pH 8.0)+ 50 mM NaCl for Q-500

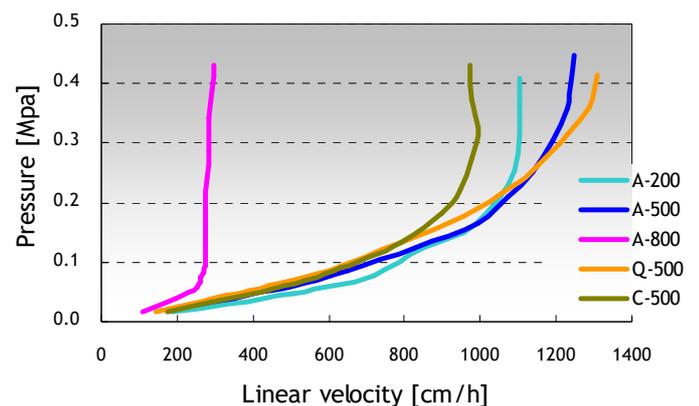


Fig 5. Pressure-flow velocity curves of Cellufine IEX media (2.2 cm I.D. x 20 cm L), Mobile phase with pure water at 24 °C,

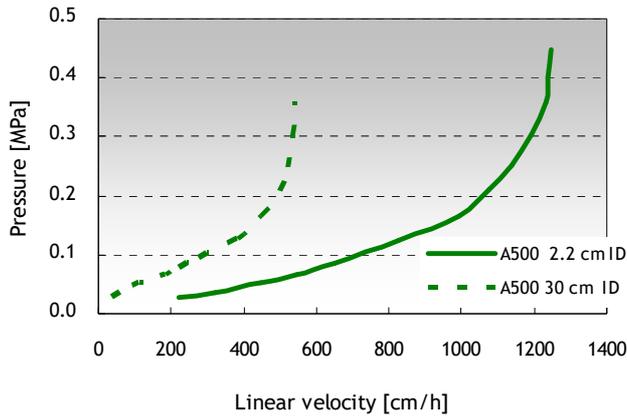


Fig 6. Pressure-flow velocity curves of Cellufine A-500 media (2.2 cm I.D. x 20 cm L and 30 cm I.D. x 20 cm L), Mobile phase with pure water at 24 °C,

Dynamic Binding Capacities of Cellufine IEX Media

Cellufine IEX media have efficient mass-transfer characteristics. Especially they exhibit superior dynamic binding performance for large molecular proteins such as immunoglobulin in comparison with competitive medium as shown in Fig. 7 and Fig.8.

These unique characteristics of Cellufine IEX media make it suitable for use in up-stream as well as to down-stream steps in bio-pharmaceuticals purification.

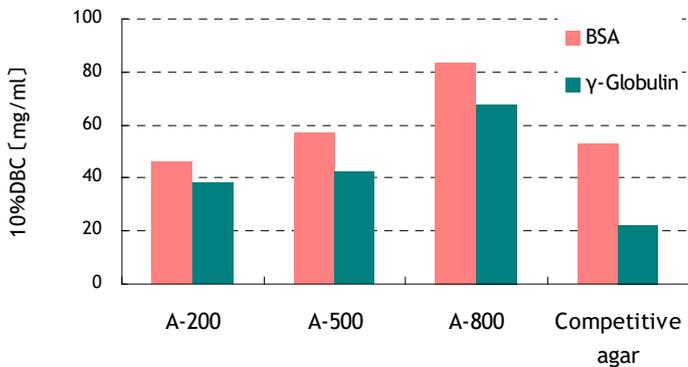


Fig. 7. DBC with model proteins in Cellufine weak anion IEX media and in competitive media

Column: 5 mm ID× 50 mm L
 Flow rate: 150 cm/h
 Sample: 1 mg/ml
 Buffer: 50 mM Tris-HCl (pH 9.5) for IgG
 50 mM Tris-HCl (pH 8.5) for BSA

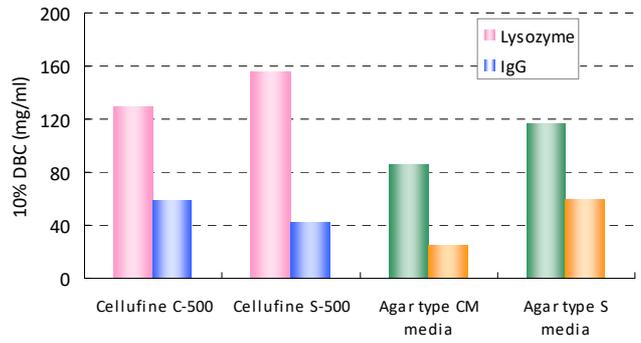


Fig. 8. DBC with model proteins in Cellufine C-500 weak cation IEX media and in competitive medium

Column: 5 mm ID× 50 mm L
 Flow rate: 150cm/h
 Sample: 1 mg/ml
 Buffer: 10 mM Acetate (pH 5.3) for IgG
 50 mM Tris-HCl (pH 8.5) for Lysozyme

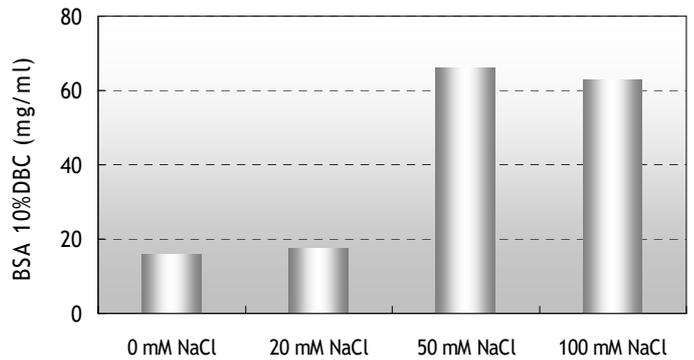


Fig. 9. DBC of BSA at different NaCl concentration for Cellufine Q-500 IEX medium

Column: 5 mm ID× 50 mm L
 Flow rate: 150 cm/h
 Sample: 1 mg/ml
 Buffer: 50 mM Tris-HCl (pH 8.5) + NaCl

Model Proteins Separation Performance for Cellufine IEX Media

Cellufine IEX media are optimized for high adsorption and high resolution. Model protein separations with Cellufine IEX media are demonstrated in Fig. 10 to 12.

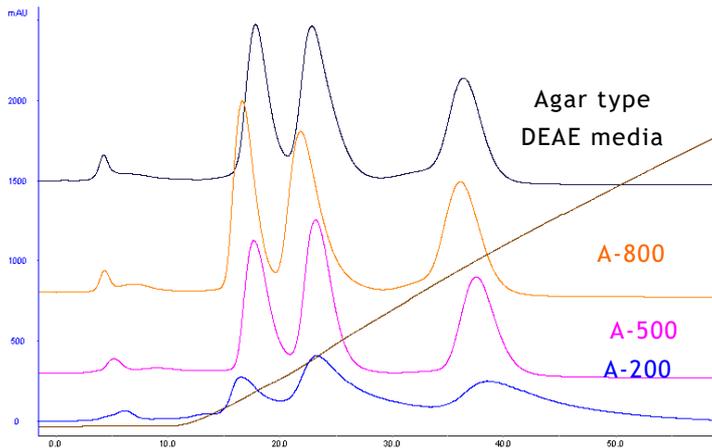


Fig. 10 Model proteins separation for Cellufine IEX media and competitive agar.

Column: 6.6 mm ID × 50 mm L

Buffer A: 50 mM Tris-HCl (pH 8.5)

Buffer B: 50 mM Tris-HCl (pH 8.5) + 1 M NaCl

(0→75 % linear gradient)

Flow rate: 0.86 ml/min (residence time 2 min)

Proteins: Transferrin (5 mg/ml),

SA (10 mg/ml),

Pepsin (5 mg/ml)

Injection volume: 1.5 ml

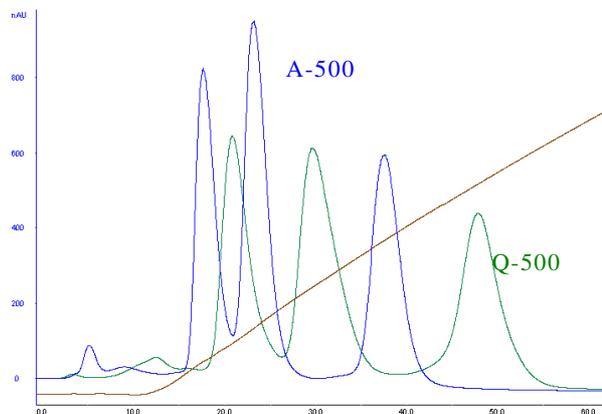


Fig. 11 shows that Model protein separation with Cellufine A-500 and Cellufine Q-500.

Column: 6.6 mm ID × 50 mm L

Buffer A: 50 mM Tris-HCl (pH 8.5)

Buffer B: 50 mM Tris-HCl (pH 8.5) + 1 M NaCl

(0→75 % linear gradient)

Flow rate: 0.86 ml/min (residence time 2 min)

Proteins: Transferrin (5 mg/ml),

BSA (10 mg/ml),

Pepsin (5 mg/ml)

Injection volume: 1.5 ml

Chemical Stability and Cleaning-In-Place

Cellulose is well-known as natural products having chemical and physical stability. Thus, since Cellufine is derived from cellulose, it also is stable to chemicals, caustic and acidic solutions. CIP of all Cellufine IEX media can be carried out with 0.5 M NaOH solution. Used media should be stored in 20 % ethanol at 2-25°C after cleaning.

Ordering Information

| Product Name | Pack Size | Catalogue No. | Product Name | Pack Size | Catalogue No. |
|--------------------|-----------------------|---------------|--------------------|-----------------------|---------------|
| Cellufine A-200 | 1ml x 5 (Mini-Column) | 19611-51 | Cellufine Q-500 | 1ml x 5 (Mini-Column) | 19907-51 |
| | 100 ml | 676980327 | | 5ml x 5 (Mini-Column) | 19907-55 |
| | 500 ml | 19611 | | 100 ml | 675982327 |
| | 5 lt | 19612 | | 500 ml | 19907 |
| | 10 lt | 676980335 | | 5 lt | 19908 |
| | | | 10 lt | 675982335 | |
| Cellufine A-500 | 1ml x 5 (Mini-Column) | 19805-51 | Cellufine C-500 | 1ml x 5 (Mini-Column) | 19800-51 |
| | 5ml x 5 (Mini-Column) | 19805-55 | | 5ml x 5 (Mini-Column) | 19800-55 |
| | 100 ml | 675980327 | | 100 ml | 675983327 |
| | 500 ml | 19805 | | 500 ml | 19865 |
| | 5 lt | 19806 | | 5 lt | 19866 |
| | 10 lt | 675980335 | 10 lt | 675983365 | |
| Cellufine A-800 | 1ml x 5 (Mini-Column) | 19865-51 | Cellufine S-500 | 1ml x 5 (Mini-Column) | 21200-51 |
| | 5ml x 5 (Mini-Column) | 19865-55 | | 5ml x 5 (Mini-Column) | 21200-55 |
| | 100 ml | 673980327 | | 100 ml | 21200 |
| | 500 ml | 19800 | | 500 ml | 21201 |
| | 5 lt | 19801 | | 5 lt | 21202 |
| | 10 lt | 673980335 | 10 lt | 21203 | |

Technical Support contacts

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