Chromatography Media

Cellufine® GH-25

Technical Data Sheet

JNC CORPORATION

Life Chemicals Division

2-1, Otemachi 2-Chome
Chiyoda-ku, Tokyo 100-8105 JAPAN
Phone +81-3-3243-6150
Fax +81-3-3243-6219
e-mail: **cellufine@jnc-corp.co.jp**
Cellufine web: **http://www.jnc-corp.co.jp/fine/en/cellufine/**

For rapid protein desalting buffer exchange and removal of alcohol and detergents

Cellufine GH-25 desalting media is based on porous, spherical, highly crosslinked cellulose particles. The sharp 3kD exclusion limit allows proteins to pass through the column in the void volume while retarding smaller molecular weight solutes in the internal pores. Outstanding mechanical strength allows operation at high flow rates even in large diameter process scale columns, thus minimizing run times.

Features

- Mechanical robust spherical particle
- Efficient salt removal
- Hydrophilic
- Pre-swollen
- pH stable 1 – 14 (0.1M HCl, 0.5M NaOH)
- Resistant to organic solvents
- Autoclavable (121 °C, 30 min)

Benefits

- Enables high flow rates and short run times
- Permits large sample loads (typical: 5 – 30 minute run times in columns from 1 ml to 100 liters, with loads up to 35 % bed volume)
- Low non-specific adsorption, high recovery
- Easy packing
- Easy cleaning and depyrogenation
- Permits use with all commonly used solvents and buffers without shrinkage or swelling
- Sterilizable

Characteristics	
Matrix	Spherical cellulose
Particle Size	ca. 40 – 130 µm
Gel Exclusion Limit	3kD
Efficiency	98 % to 100 % recovery. No deterioration after 250 days, 1000 cycles.
Autoclavable	121 °C, 30 min.
Pressure Resistance	No collapse at up to 870 ml/h/cm^2 flow in large columns
pH Stability	pH 1 – 14
Chemical Resistance	Resistance to detergents and dissociating agents. No change after 30 days in 0.1M HCl or 0.1M NaOH.
Supplied	Suspension in 20 % ethanol

Flow Properties

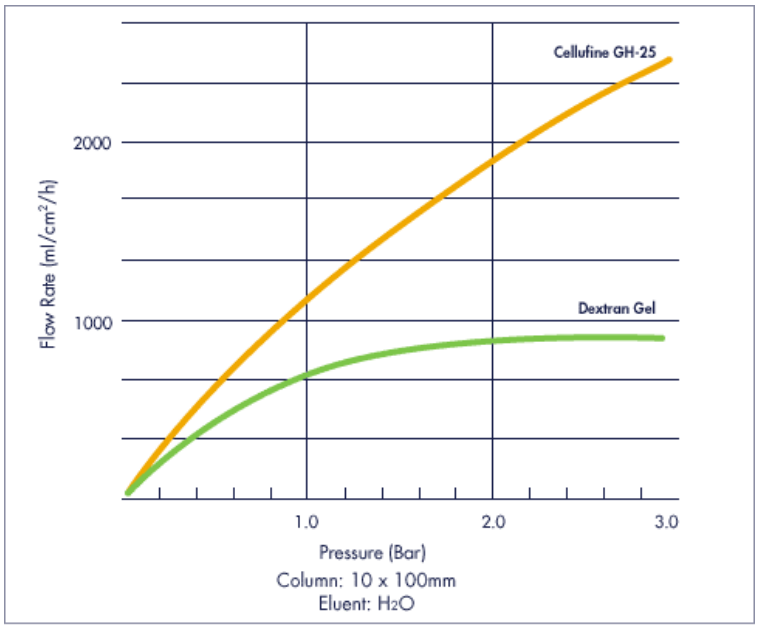
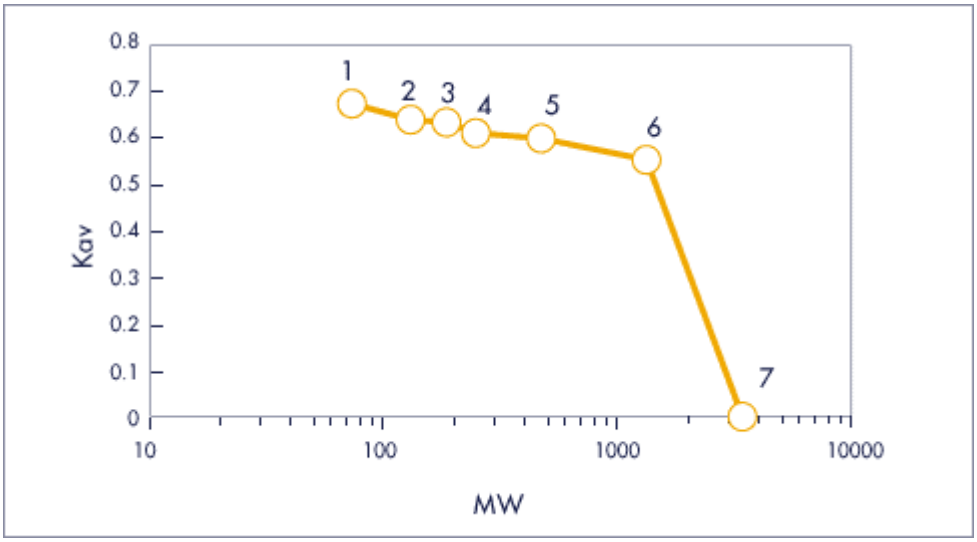


Figure 1
Pressure/flow characteristics of Cellufine GH-25 vs Dextran gel

Due to its rigidity, Cellufine GH-25 delivers nearly twice the flow rate of an equivalent sized Dextran gel.

The specific selection curve of Cellufine GH-25



1: Glycine 75 **2**: $(Gly)_2$ 132 **3**: $(Gly)_3$ 189 **4**: $(Gly)_4$ 246
5: Calcium pantothenate 477 **6**: Vitamin B12 1355 **7**: insulin B chain 3495

Applications

- Desalting before lyophilization or concentration
- Buffer exchanges
- Removal of alcohol or other organic solvents
- Removal of aromatic compounds (e.g., phenol) in purification of nucleic acids
- Removal of detergents used to solubilize proteins (e.g. Triton® X-100, SDS)
- Permits use with all commonly used solvents and buffers without shrinkage or swelling
- Removal of chaotropic agents, (e.g. urea, guanidine)

High Speed Desalting

Although the rigidity of Cellufine GH-25 makes it ideally suited to large scale column use, its ability to operate at very high flow rates enables the use of smaller columns running multiple cycles giving similar throughput to lower flow rates on larger columns. Unlike conventional chromatography, the performance (as measured by sample load, salt removal, and dilution) of desalting chromatography can actually improve as the flow rate increases, due to decreased sample dilution at high volumetric loads.

Protein Desalting

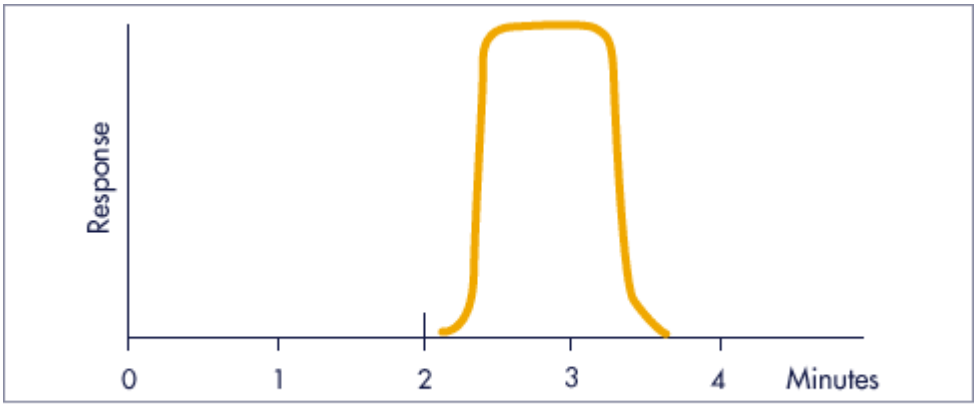


Figure 2
High speed protein desalting

Packing :	Cellufine GH-25
Column :	105 x 587 mm Vt = 5086 ml
Mobile Phase :	0.1M NaCl
Flow :	1250 ml/min, 870 ml/h/cm^2
Sample :	5 % (w/v BSA) in 1.5M NaCl
Sample Volume :	1272 ml (25 % of column volume)
Protein Recovery :	99.5 %
Salt Exchange :	99 %
Dilution :	1.13x

Alcohol Removal

Fractionation of human blood and the subsequent removal of alcohol from the albumin are two important elements of albumin production. Table 1 compares alcohol removal from albumin with GH-25 gel at two different flow rates. Increasing the flow rate does not affect de-alcoholization efficiency, albumin recovery or sample dilution. Concentration of the remaining alcohol was below 0.01 % for either flow rate. Cycle time was reduced to one quarter by increasing flow from 29 cm/h/cm² to 100 cm/h/cm² .

Run Number	1	2
Media type	Cellufine GH-25	
Column diameter (mm)	50	50
Column diameter length (mm)	680	670
Media volume (ml)	1335	1320
Flow rate (ml/hr)	570	2010
Linear velocity (ml/h/cm²)	29	102
Time needed for a cycle (hr)	2.3	0.6
Product Applied		
Process volume (ml)	310	310
Process volume (% Vt)	23	23
Albumin concentration (%)	12	12
Ethanol concentration (%)	4.8	4.8
Product Collected		
Recovered volume of product (ml)	546	525
Dilution factor	1.8x	1.7x
Recovered albumin concentration (%)	6.6	6.9
Remaining alcohol concentration (%)	0.002	0.01
Mass recovery of albumin (%)	97	98

Table 1. Alcohol removal from human albumin fractionation procedure

Industrial Desalting

The advantages of desalting biological molecules chromatographically are clearly realized in large-scale applications. From research to pilot and production facility, the scale-up of Cellufine GH-25 has proven to be direct and trouble-free. Mechanical stability of GH-25 allows the use of high flow rates in large columns.

A typical large scale desalting application requiring the processing of 225 liters per day can be accomplished by 5 liters of Cellufine GH-25 in a 105 x 587 mm column (see Table 2). This rigid gel withstands flow rates to 1250 ml/min in that column geometry (870 ml/cm/cm^2). Cycle time between injections is 8 minutes. The high volume load (25 % of Vt), is accommodated by the packing without sacrificing resolution or purity.

Packing	Cellufine GH-25
Column (mm)	105 x 587
Vt	5.1 liters
Flow rate (ml/min) (liters/hr)	1,250 (75 liters/hr)
Linear velocity	870 ml/h/cm^2
Product	5 % w/v protein in 1.5M NaCL
Product volume	1.27 liters (25 % of Vt)
Product mass/cycle	63.6g
Cycles/day	180
Product mass/day	11.4kg
Protein mass recovery	99.5 %
Salt exchange	99 % (0.015M final conc.)
Sample dilution	1.13
Total product volume recovered	257 liters
Total product volume processed per day	225 liters

Table 2. Throughput analysis for high speed desalting

Ordering Information

Product Name	Pack Size	Catalogue No.
Cellufine GH-25	5ml x 5 (packed column)	19711-55
	100 ml	670 000 327
	500 ml	19711
	5 Liters	19712
	10 Liters	670 000 335

Contact us

(Technical Support)

(North America & Europe)
JNC America Incorporated
555 Theodore Fremd Avenue, Suite C-206
Rye, NY 10580 USA
TEL: 914-921-5400
FAX: 914-921-8822
E-mail: cellufine@jncamericany.com

(Asia & Others)
JNC Corporation
Life Chemicals Division
2-1, Otemachi 2-Chome, Chiyoda-ku
Tokyo 100-8105 Japan
Tel: +81-3-3243-6150
Fax: +81-3-3243-6219
E-mail: cellufine@jnc-corp.co.jp

(Purchase Orders)

(North America & Europe)
JNC America Incorporated
555 Theodore Fremd Avenue, Suite C-206
Rye, NY 10580 USA
TEL: 914-921-5400
FAX: 914-921-8822
E-mail: cellufine@jncamericany.com

(Asia & Others)
JNC Corporation
Life Chemicals Division
2-1, Otemachi 2-Chome, Chiyoda-ku
Tokyo 100-8105 Japan
Tel: +81-3-3243-6150
Fax: +81-3-3243-6219
E-mail: cellufine@jnc-corp.co.jp