Cellufine™
Purification Solutions

Virus Purification, mAb Polishing, and mRNA-related Enzyme Purification

As clinical pipelines increase for biopharmaceuticals, viral vaccines, and mRNA-based vaccines, the need to purify these emerging products requires new and innovative purification solutions. Cellufine chromatography media is designed for the purification of therapeutic proteins, enzymes, viruses, and other biomolecules.

Based on porous, spherical cellulose beads, Cellufine is inherently biocompatible and exhibits significantly lower leachables than synthetic polymers, minimizing potential contaminants. Its high chemical stability ensures a long media lifetime by withstanding repeated and aggressive cleaning-in-place (CIP) cycles, to reduce overall costs and maximize process uptime. These combined advantages make Cellufine the ideal choice for the robust purification of today's most demanding biomolecules.

In the following purification cases, we will explore how various Cellufine chromatography medias can improve your purity and throughput for virus purification, mAb polishing, and mRNA-related enzyme purification, respectively.

PURCHASING / TECHNICAL SUPPORT

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Virus & Viral Vaccine Purification

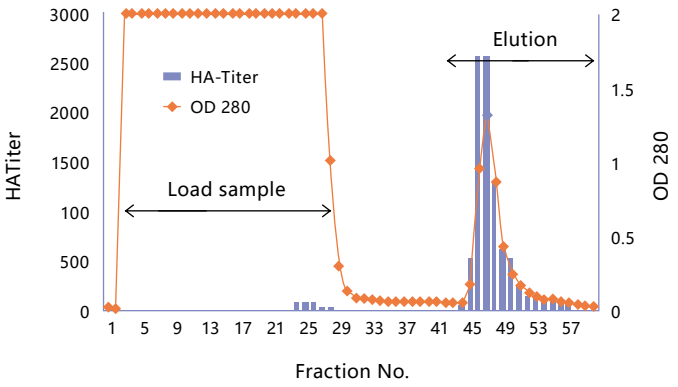
Cellufine™ Sulfate

Fast virus purification with no pre-treatment, high recovery, and impurity removal with proven performance.

Egg-Cultured Influenza Virus (H7N7)[1]

In Figure A, we demonstrate the ability of Cellufine Sulfate to purify H7N7 egg-cultured influenza virus.

Figure A: Egg-cultured influenza virus (H7N7)



Step	HA Titier	Protein (μg)
Load Sample	102,400	9,848
Elution	91,520 (89%)	800 (8%)

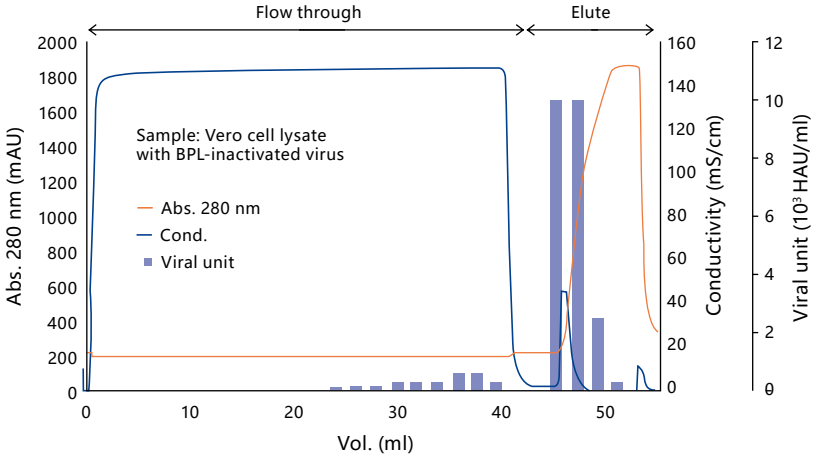
Conditions

Column: 8 mm ID x 20 mm L (1 mL)
Linear Velocity: 120 cm/hr
Equilibration buffer: 0.01 M Phosphate buffer (pH 7.4)
Washing buffer: 0.01 M Phosphate buffer, 0.19 M NaCl (pH 7.2)
Elution buffer: 0.01 M Phosphate buffer, 3 M NaCl (pH 7.0)
Virus Strain: A/duck/Hokkaido /Vac-2/04 (H7N7) treated with β-Propiolactone
Sample Volume: 40 mL

Human Coronavirus (OC43)[2]

In Figure B, we show purification of OC43 human coronavirus. Coronavirus has an envelope protein.

Figure B: Purification of human coronavirus (OC43) with Cellufine Sulfate



Recovery (%)	Dynamic Binding Capacity (HA/ML resin)*
77	76,200

* Measured at 10% breakthrough point

Conditions

Column: 5 mm ID x 15 mm L (0.3 mL)
Flow rate: 0.3 mL/min (residence 1 min)
Equilibration: 10 mM Na_3PO_4, 150 mM NaCl (pH 7.4)
Elution: 10 mM Na_3PO_4, 2 M NaCl (pH 7.4)

Protein Purification

Beyond viruses, this media supports protein purification with heparin-like affinity and capacity similar to heparin resins.

Binding Proteins	
Antithrombin III	Chymotrypsinogen
β-Lipoprotein	Lysozyme
Complement C5, C6, C8	Urease
Complement C3 Activator	Catalase
Trypsin	Factor IX
Trypsin Inhibitor	

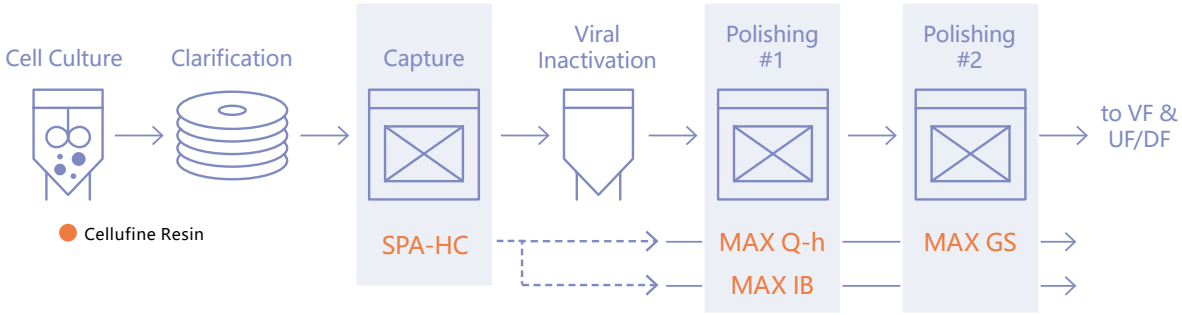
Non-Binding Proteins	
Albumin	Ceruloplasmin
α-Lipoprotein	α2-Macroglobulin
Complement C3, C9	RNase
Complement C1, C3b	Bacitracin
Inactivators	Glucose Oxidase
IgG	

[1] Y. Sakoda *et al.* Purification of human and avian influenza viruses using cellulose sulfate ester (Cellufine Sulfate) in the process of vaccine production. *Microbiology and Immunology*, 56, 490–495 (2012).
[2] Kenji Kadoi *et al.* Enhanced sulfate pseudo-affinity chromatography using monolith-like particle architecture for purifying SARS-CoV-2. *Vaccine*, 53, 126951 (2025).

Monoclonal Antibody (mAb) Polishing

mAb Process

As antibody purification becomes more demanding, new tools are needed in the downstream polishing toolbox. Below, we show two polishing approaches for mAbs using flow-through (FT) and bind-and-elute (B/E) modes.



Case 01: Cellufine™ MAX Q-h & MAX GS

High antibody recovery rate; significant reduction of HCP* + aggregates

Cellufine MAX Q-h is a highly crosslinked strong anion exchange resin with high binding capacity; Cellufine MAX GS is a novel strong cation exchange resin designed specifically for removal of mAb aggregates. Together, they provide a powerful polishing workflow for mAbs as shown in the table to the right.

*HCP: Host Cell Protein

Media	mAb Load (mg/mL)	HCP (ppm)	Leached ProteinA (ppm)	Aggregates (%)	Recovery (%)
Supernatant		928,000			
SPA-HC (Protein A)	53	2350	14.2	1.8	95
MAX Q-h (FT)	185	49	8.0	1.2	98
MAX GS (B/E)	58	5	0.2	0.5	90

Conditions of MAX GS

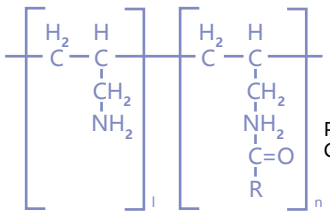
Sample: mAb solution after MAX Q-h, 6.04 mg/mL, 6 mS/cm (pH 5.0)
Column: Super Edge™ 1 mL (6.7 mm ID x 30 mm L, CV = 1.06 mL, JNC) Resin; Cellufine MAX GS
Load Amount: ~ 60 mg-mAb/mL-resin
Eq. and Wash Buffer: 20 mM Acetate + 50 mM NaCl (pH 5.0)
Elution Buffer: 20 mM Acetate-Na + 0.19 M NaCl (pH 5.0)

Case 02: Cellufine™ MAX IB

Reduction of purification step and excellent removal of HCP + aggregates

Our mixed mode resin, Cellufine MAX IB, has a salt tolerant polyamine surface modification that has been partially modified with butyl groups. Its novel ligand design provides the possibility of single-step mAb polishing after Protein A capture, as demonstrated here.

Ligand structure



Partial structure of Cellufine MAX IB

	HCP (ng/mg mAb)	Aggregates (HPLC %)		mAb yield (%)
		HCP (ppm)	HMWs	
Load	428	1.49	0.26	100
Capto™ adhere	4	0.92	Not detected	92
MAX IB (FT)	2	0.87	Not detected	88

Conditions**

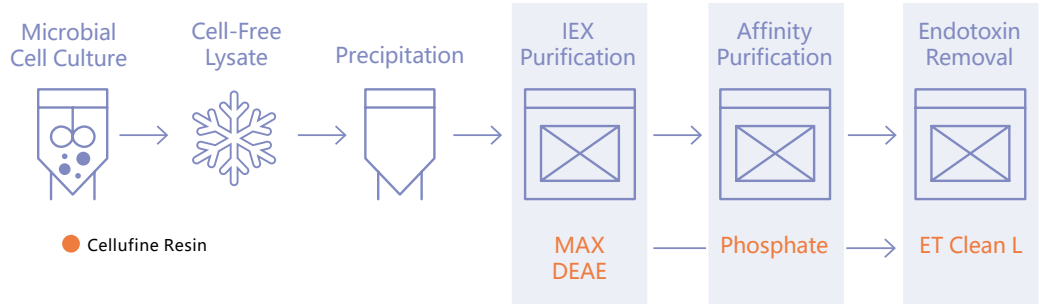
Column: Tricorn 5/50; 5 mm ID x 50 mm L (0.98 mL, Cytiva)
Buffer: 20 mM Acetate-Tris (pH 7.0), NaCl, 6 mS/cm
Sample: 11.7 mg/mL partially purified mAb, 25 mL
Load Amount: 293 mg-mAb/mL-resin
Flow Rate: 0.25 mL/min (residence time: 4 min)

**mAb1 CHO cells (pI: 8-9) were eluted with Protein A using 60 mM acetate buffer (pH 3.5). Virus inactivation to pH 3.4, adjusted by 0.1 M HCl at 25° C for 1 hour prior to mixed mode polishing.

mRNA-Related Enzyme Purification

T7 RNA Polymerase Process

In vitro transcription (IVT) for mRNA production utilizes enzymes such as T7 RNA polymerase to synthesize RNA from a DNA template. We have developed a range of chromatography resins that are ideal for the purification of these nucleic acid-related enzymes.



Cellufine™ Phosphate

Capable of highly purifying enzymes

Figure C: Purification of T7 RNA polymerase with Cellufine Phosphate

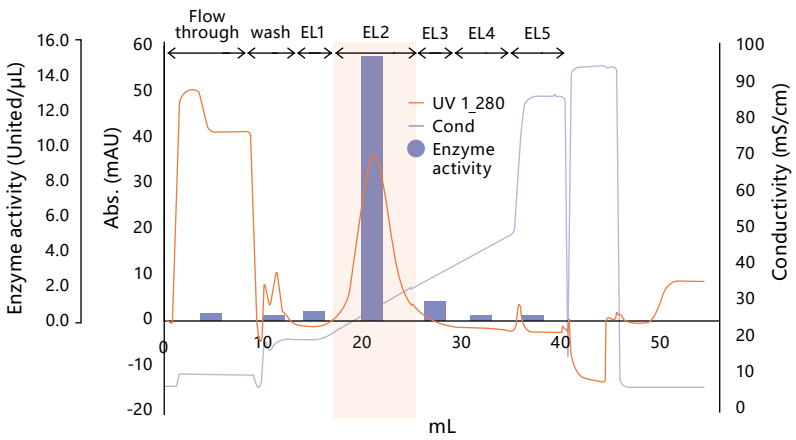
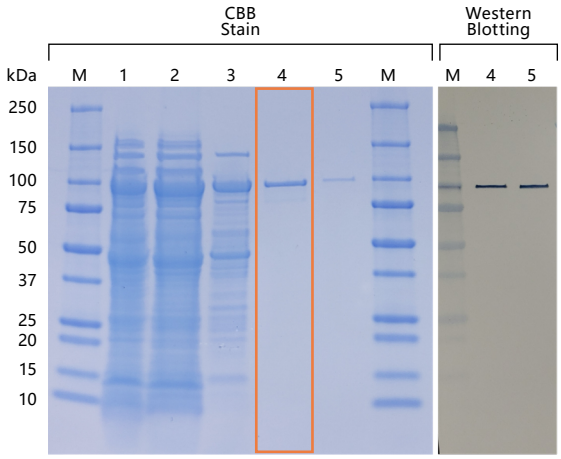


Figure D: SDS-PAGE of Phosphate fractions



Conditions

Sample: post ammonium sulfate precipitation and MAX DEAE purification (adjusted 10 mS/cm) of cell culture derived from *E. coli.*

Column: Super Edge™ 1 mL (6.7 mm ID x 30 mm L, CV = 1.06 mL, JNC)

Flow rate: 0.5 mL/min (residence time 2 min)

Loading and wash buffer: 10 mM potassium phosphate (pH 7.5), 50 mM NaCl, 0.1 mM EDTA, 0.1 mM DTT, protease inhibitor

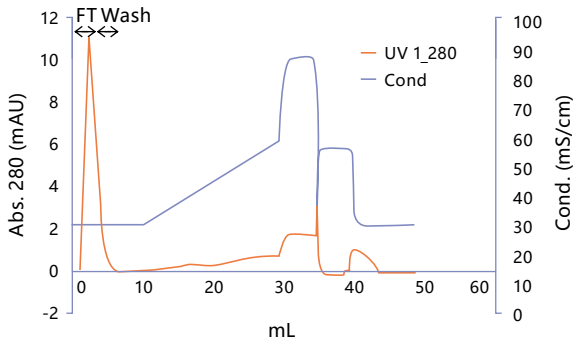
Elution buffer: 1 M NaCl in loading buffer

M: Marker
1: Lysate
2. $(NH_4)_2SO_4$ precipitate
3. Cellufine MAX DEAE Elution
4: Cellufine Phosphate Elution
5. Positive Control (PC)

Cellufine™ ET clean L

Capable of removing endotoxins derived from *E. coli.*

Figure E: Endotoxin removal from T7 RNAP



Step	T7 RNAP (U/mL)	Endotoxin (EU/μg)	Endotoxin Reduction (%)	Endotoxin (EU/unit T7 RNAP)	Endotoxin Protein (EU/μg)
Load Sample	12140	134	–	11.0	3.46
FT + Wash	5360	0.23	99.6	0.04	0.01

Conditions

Sample: Diafiltered Phosphate elution fraction 2

Column: Super Edge™ 1 mL (6.7 mm ID x 30 mm L, CV = 1.06 mL, JNC)

Flow Rate: 0.5 mL/min (residence time: 2 min)

Load Buffer: 10 mM Tris-HCl (pH 7.5), 300 mM NaCl, 0.1 mM EDTA, 0.1 mM DTT

High Salt Buffer: 10 mM Tris-HCl (pH 7.5), 1 M NaCl, 0.1 mM EDTA, 0.1 mM DTT