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A Geno Technology, Inc. (USA) brand name

G-Sep™ Agarose Fast Flow

(Cat. # 786-954)

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INTRODUCTION

G-Sep™ Agarose 6 Fast Flow (FF) is a gel filtration matrix formed from agarose beads that is based on cross-linked 6% agarose.

G-Sep™ Agarose CL resins are cross-linked versions of our G-Sep™ Agarose. Cross-linking of the agarose results in chemically and physically more stable agarose beads that offer the same selectivity, but with better flow characteristics. Cross-linked agarose beads are resistant to organic solvent.

The modification to the cross-linked 6% agarose to fast flow results in improved physical stability and chromatographic qualities. The modification makes the resin an ideal base resin for high throughput applications and industrial process separations. The improved rigidity permits higher flow rates resulting in improved resolution in minimum time. G-Sep™ Agarose 6 FF can also be used for the immobilization of ligands for improved affinity chromatography.

ITEM(S) SUPPLIED

Cat. #	Description	Size
786-954	G-Sep™ Agarose 6 Fast Flow	1L

STORAGE CONDITIONS

It is shipped at ambient temperature. Upon arrival, store at 4°C. DO NOT FREEZE.

SPECIFICATIONS

	6-FF
Matrix	Highly cross-linked Agarose, 6%
Bead form	Spherical, diameter 50-160µm
pH stability Working Range	3-13
pH stability Cleaning-in-Place (CIP)	2-14
Maximum Pressure (MPa)	0.3
Maximum Flow Velocity	450cm/h
Fractionation [Mr] Globular Proteins	1×10^4-4×10^6
Physical Stability	Negligible volume variation due to changes in pH or ionic strength
Chemical Stability	Stable to: 6M urea, 8M guanidine hydrochloride, ethanol, DMF, THF, acetone, DMS, chloroform, dichloromethane, dichloroethane, pyridine, triethyl phosphate and acetonitrile.
Sterilization	Autoclavable, 121°C, pH 7, for 20 min
Storage Conditions	4 to 30°C, 20% Ethanol

PREPARING THE MEDIUM

G-Sep™ Agarose 6 Fast Flow is supplied in a solution containing 20% ethanol.

Prepare a 75% slurry using a binding/ eluent buffer of choice. The slurry will be 75% settled resin and 25% buffer. It is recommended to degas the slurry before packing.

PACKING G-SEP™ AGAROSE FAST FLOW

1. Equilibrate all material to room temperature.
2. De-gas the slurry
3. Eliminate air from the column dead spaces by flushing the end pieces with buffer. Make sure no air has been trapped under the column net. Close the column outlet with a few centimeters of buffer remaining in the column.
4. Pour the gel slurry into the column in one continuous motion. Pouring the slurry down a glass rod held against the wall of the column will minimize the introduction of air bubbles.
5. Fill the remainder of the column with buffer, mount the column top piece onto the column and connect the column to a pump.
6. Open the bottom outlet of the column and set the pump to run at the desired flow rate. This should be at least 133% of the flow rate to be used during subsequent

chromatographic procedures. However, the maximum flow rate is typically employed during packing.

7. Maintain the packing flow rate for 3 bed volumes after a constant bed height is reached.

Using an adapter

1. After the medium have been packed as described above, close the column outlet and remove the top piece from the column. Carefully fill the rest of the column with buffer to form an upward meniscus at the top.
2. Insert the adaptor into the top of the column at an angle, taking care not to trap air under the net.
3. Make all tubing connections at this stage. There must be a bubble-free liquid connection between the column and the pump.
4. Slide the plunger slowly down the column so that the air above the net and in the capillary tubings is displaced by eluent. Valves on the inlet side of the column should be turned in all directions during this procedure to ensure that air is removed.
5. Lock the adapter in position on the medium surface, open the column outlet and start the eluent flow. Pass eluent through the column at the packing flow rate until the bed is stable. Re-position the adapter on the medium surface as necessary.
6. The column is now packed and equilibrated and ready for use.

OPERATION

Equilibration

Equilibrate the column with the starting buffer when the pH and/or conductivity of the effluent is the same as the starting buffer.

Sample preparation

Before application the sample should be centrifuged or filtered through a 0.45µm filter to remove any particulate matter. Recommended sample volume is 2-5% of the total bed volume.

Elution

It is recommended to use a buffer with an ionic strength of 0.15 or greater to avoid any unwanted ionic interactions between the solute molecule and the agarose beads.

Regeneration

After every run, elute reversibly bound material with low ionic strength buffer, and wash with H_2O and starting buffer.

Cleaning-in-place(CIP)

Remove precipitated proteins and hydrophobically bound proteins or lipoproteins: Wash with 1-2M NaOH and immediately rinse with eluent buffer.

Lipids and very hydrophobic proteins: Wash with 70% ethanol, reversed flow for 1-2 hours. Alternatively wash with saw-tooth gradients of 0-30% isopropanol. Contact time 1–2 hours.

After cleaning, equilibrate the column with sterile start buffer before use. Alternatively, wash the column with detergent in a basic or acidic solution. Use e.g. 0.5% non-ionic detergent in 1M acetic acid. Remove residual detergent by washing with 70% ethanol.

Sanitization

Wash the column with 1-2M NaOH for 30-60 min. Sanitization is the use of chemical agents to inactivate microbial contaminants in the form of vegetative cells; it also helps to maintain a high level of both process hygiene and process economy.

Storage

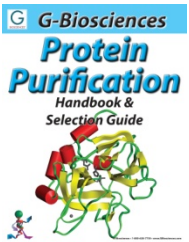
G-Sep™ Agarose Fast Flow should be stored in the salt form in a buffer containing 20% ethanol. Recommended storage at 4 to 30°C. Do not freeze.

Shelf life

5 year

RELATED PRODUCTS

Download our Sample Preparation and Protein Purification Handbooks.



http://info2.gbiosciences.com/complete-protein-purification-handbook

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