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PreCap LCA

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1. Product Description

LCA Beads 4FF is an affinity chromatography medium that conjugates Lens Culinaris Agglutinin (LCA) with agarose to purify some glycoproteins. LCA is a metalloprotein separated from lentils, which can binds α-D- pyran mannose, α-D- pyran on glucose and Molecular groups related to its space position of the combination. **LCA Beads 4FF** is used to isolate and purify some glycoproteins, membrane proteins, glycolipids, polysaccharides, membrane vesicles with mannoside or glucoside residues, IgM, hormone lipoproteins and so on. LCA Beads 4FF has a wide range of applications, and the characteristics are shown in Table 1.

PreCap LCA is one of a range of prepacked, ready-to-use columns for affinity chromatography. It is packed with 1 ml and 5 ml of LCA Beads 4FF. Five different packing sizes are available. PreCap LCA has the standard interface and can be adapted to all kinds of chromatography system, such as ÄKTA. It is fast, simple and easy operation.

Table 1. Characteristics of **LCA Beads 4FF**

Item	Description
Matrix	Highly cross-linked 4% agarose
Ligand	Lens Culinaris Agglutinin
Binding capacity	>13 mg thyroglobulin /ml medium
Particle size (μm)	45-165
Maxi pressure	0.3 MPa, 3 bar
Storage buffer	150 mM NaCl, 1 mM $CaCl_2$, 1 mM $MnCl_2$, 20% ethanol
Storage	2-8℃

2.Purification Procedure

2.1 Buffer Preparation

It is recommended to filter the buffers by passing them through a 0.22 μm or 0.45 μm filter before use.

Mn^{2+} and Ca^{2+} must be added to ensure adsorption activity when the buffer pH is lower than 5.0. We recommend using the following buffer for purification.

Binding /Wash Buffer: 20 mM Tris-HCl, 0-0.5 M NaCl, 1 mM $CaCl_2$, 1 mM $MnCl_2$, pH 7.4

Elution Buffer: 20 mM Tris-HCl, 0.5 M NaCl, 1 mM $CaCl_2$, 1 mM $MnCl_2$, 0.1 M-0.2 M Dew α-D- methyl glucoside or α-D- methyl glucoside, pH 7.4

Note: In the eluent Dew α-D- methyl glucoside or α-D- methyl glucoside concentration according to physical adsorption capacity for linear or gradient elution. Mannose and glucose can also be used as eluent, but the eluent ability is weak.

For substances with strong binding capacity, the pH of the lower eluent can be eluted, but not lower than pH 3.0.

Borate can be used as eluent, such as 0.1 M borate, pH 6.5.

For strongly bound proteins, 1% sodium deoxycholate or other detergent can be added to the elution buffer to facilitate elution.

2.2 Sample Preparation

To ensure that the sample solution has the appropriate ionic strength and pH before ascending the column, dilute the serum sample, ascites, or cell culture with a balance/wash mixture, or dialysis the sample with a balance/wash Buffer.

Samples should be centrifuged or filtered by a 0.22 μm or 0.45 μm membrane before loading to reduce impurities, improve protein purification efficiency and prevent clogging of the column.

2.3 Sample Purification

1) Fill the syringe or pump tubing with distilled water. Remove the stopper and connect the column to the syringe (with the provided connector), or pump tubing, “drop to drop” to avoid introducing air into the column. Remove the stopper at the column outlet and connect the column to the chromatographic system.

2) Wash the column with 3-5 column volumes of distilled water.

3) Equilibrate the column with at least 5 column volumes Lysis Buffer.

4) Apply the pre-treated sample, using a Loop fitted to the connector or by pumping it onto the column.

5) Wash with Wash Buffer until the absorbance reaches the baseline or no material appears in the effluent (Generally at least 10-15 column volumes).

6) Elute with 5 column volumes of elution buffer. Other volumes may be required if the interaction is difficult to break.

2.4 Analysis

SDS-PAGE will be used to test the purification effect of the samples obtained from the purified product (including the effluents, eluted and eluted components) and the original samples.

3. Cleaning-In-Place

In general, **LCA Beads 4FF** is well suited for reuse a number of times. When precipitation and protein aggregation cause the loss of velocity and combined loads, you need to clean the medium as follows.

Wash with 3-4 column volumes 0.5 M NaCl, pH 8.5 and 0.5 M NaCl, pH 4.5 for 3 times alternately, then balance with 5 column volume binding buffer.

Remove the strong binding proteins

Wash with 2 column volumes of 0.1 M borate buffer containing 1% Triton™ X-100 with pH 6.5, or 20% ethanol, or 50% ethylene glycol, then immediately balance with 5 column volume binding buffer.

4. Related Products

Product	Cat. No.	Size
LCA Beads 4FF	SA097005	5 ml
	SA097025	25 ml
	SA097100	100 ml
	SA097500	500 ml
	SA09701L	1 L
PreCap LCA	SA097C11	1×1 ml
	SA097C51	5×1 ml
	SA097C15	1×5 ml
	SA097C55	5×5 ml
	SA097CS	3×1 ml+1×5 ml