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PreCap Butyl 4FF
PreCap Octyl 4FF
PreCap Phenyl LS 6FF
PreCap Phenyl HS 6FF

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1. Product Description

Butyl Beads 4FF, Octyl Beads 4FF, Phenyl Beads 6FF(Low Sub) and Phenyl Beads 6FF(High Sub) are media for Hydrophobic Interaction Chromatography(HIC). Substances are separated on the basis of their varying strength of their hydrophobic interaction with hydrophobic groups attached to the uncharged matrix. They are developed and supported for process scale chromatography. The base matrix of **Butyl Beads 4FF, Octyl Beads 4FF, Phenyl Beads 6FF(Low Sub) and Phenyl Beads 6FF(High Sub)** is highly cross-linked agarose. They have high chemical and physical stability. Details see table under each respective HIC media.

PreCap HIC is one of a range of prepacked, ready-to-use columns for affinity chromatography. It is packed with 1ml and 5ml of HIC medium. PreCap HIC has the standard interface and can be adapted to all kinds of chromatography system, such as ÄKTA. It is fast, simple and easy operation.

Butyl Beads 4FF

Butyl Beads 4FF is aliphatic hydrophobic interaction medium. The butyl group is coupled to beads by ether linkage, giving a hydrophobic medium with minimal leakage and no ionic properties.

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

Item	Description
Matrix	Highly cross-linked 4% agarose
Type of ligand	Butyl
Capacity (/ml mediun)	approx.7 mg IgG，＞26 mg BSA
Particle Size (μm)	45-165
Flow rate	300 cm/h
pH stability	3-13
Storage buffer	20% ethanol
Storage	4℃ - 30℃

Octyl Beads 4FF

Octyl Beads 4FF is aliphatic hydrophobic Interaction medium. The octyl group is coupled to beads by ether linkage, giving a hydrophobic medium with minimal leakage and no ionic properties.

Table 2. Characteristics of **Octyl Beads 4FF**

Item	Description
Matrix	Highly cross-linked 4% agarose
Type of ligand	Octyl
Capacity (/ml medium)	approx.26 mg IgG, ＞7 mg BSA
Particle Size (μm)	45-165
Flow rate	300 cm/h
pH stability	3-13
Storage buffer	20% ethanol
Storage	4℃ - 30℃

Phenyl Beads 6FF(Low Sub) and **Phenyl Beads 6FF(High Sub)** consist of 90 μm beads of 6% highly cross-linked agarose. The phenyl group is coupled to beads by ether linkage, giving a hydrophobic medium with minimal leakage and no ionic properties. According to the required separation selectivity, efficiency and binding capacity.Different substituted medium are available.

Table 3. Characteristics of **Phenyl Beads 6FF(Low Sub)** and **Phenyl Beads 6FF(High Sub)**

Item	Description
Matrix	Highly cross-linked 6% agarose
Type of ligand	Phenyl
Capacity (/ml medium)	Low Sub:approx.10 mg IgG, ＞24 mg BSA High Sub:approx.30 mg IgG, ＞36 mg BSA
Particle Size (μm)	45-165
Flow rate	300-600 cm/h
pH stability	3-13
Storage buffer	20% ethanol
Storage	4℃ - 30℃

2. Purification Procedure

2.1 Buffer Preparation

Water and chemicals used for buffer preparation should be high purity. It is recommended to filter the buffers by passing them through a 0.22μm or 0.45μm filter before use.

Binding/wash buffer: 0.05 M phosphate, 1.7 M $(NH_4)_2SO_4$, pH7.0

Elution Buffer: 0.05 M phosphate, pH7.0

Note: The buffer of HIC can be changed according to different samples and the medium.The salt concentration of buffer is high in Binding/wash buffer and low in elution buffer.

2.2 Sample Preparation

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

The salt concentration in the sample is the same as binding /wash buffer. It is usually 0.5-2.0 M $(NH_4)_2SO_4$.

2.3 Sample Purification

1) Fill the syringe or pump tubing with binding buffer. 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 snap-off end at the column outlet.

2) Wash the column with 10 column volumes of binding buffer.

3) Apply the sample, using a syringe fitted to the connector or by pumping it onto the column.

Note:Hydrophobic interaction is weaker at lower temperatures. This must be taken into account if chromatography is done in a cold room.

4) Wash with 5 to 10 column volumes of binding buffer or until no material appears in the effluent.

5) Elute with Elution Buffer using a stepwise or linear gradient. For one-step elution, 5 column volumes are usually enough. Other volumes may be required if the interaction is difficult to break. Linear gradient elution can be used to separate proteins of different binding strengths with a small gradient, such as 20 column volumes or more.

2.4 Analysis

Identify the fractions containing the target protein. using UV absorbance, SDS-PAGE, or western blot.

3. Clean-in-Place

After each separation, elute reversibly bound material with low ionic strength buffer. Wash the column wit 5 column volumes of distilled water and 30%isopropanol.

- **Remove strongly hydrophobically bound proteins, lipoproteins and lipids**

Wash the column with 3 column volumes of 70% ethanol or 30% isopropanol (apply increasing concentration gradients to avoid air bubbles formation) . Alternatively, wash the column with 3 column volumes of 0.1-0.5% detergent in a basic or acidic solution. For example, wash with 0.1-0.5% non-ionic detergent in 0.1 M acetic acid .Contact time 1-2 h.

Wash the column with distilled water and re-equilibrate.

- **Sanitization reduces microbial contamination**

Wash the column with 1 M NaOH. Contact time 30-60 min.

Wash the column with distilled water and re-equilibrate.

4. Related Products

Product	Cat. No.	Size
PreCap Butyl 4FF	SH001C11	1X1 ml
	SH001C51	5X1 ml
	SH001C15	1X5 ml
	SH001C55	5X5 ml
PreCap Octyl 4FF	SH003C11	1X1 ml
	SH003C51	5X1 ml
	SH003C15	1X5 ml
	SH003C55	5X5 ml
PreCap Phenyl LS 6FF	SH004C11	1X1 ml
	SH004C51	5X1 ml
	SH004C15	1X5 ml
	SH004C55	5X5 ml
PreCap Phenyl HS 6FF	SH006C11	1X1 ml
	SH006C51	5X1 ml
	SH006C15	1X5 ml
	SH006C55	5X5 ml