Butyl-S Sepharose 6 Fast Flow

Product Information

Cat#No#	Bu-414C

Product Overview

Butyl-S Sepharose 6 Fast Flow is the least hydrophobic resin in the Sepharose Fast Flow hydrophobic interaction chromatography (HIC) range and is particularly useful during the initial stages of a purification process to remove the bulk of impurities.

Description

Butyl-S Sepharose 6 Fast Flow belongs to the family of products referred to as hydrophobic interaction chromatography (HIC) media. Cytiva produces a wide range of HIC media that is well characterized for group separation or purification of a variety of biological macromolecules in laboratory- or process-scale operations. Butyl-S Sepharose 6 Fast Flow is the least hydrophobic medium in the series. Butyl-S Sepharose 6 Fast Flow is particularly useful during the initial stages of a separation process to remove the bulk of impurities without stringent requirements for conditioning of the sample It is a BioProcess medium that meets the demands of large-scale biopharmaceutical manufacturers for efficient and cost-effective protein purification.

Characteristic

Designed for the binding and elution of relatively strong hydrophobic molecules at comparatively low salt concentrations.
Used in purification of recombinant Hepatitis B virus surface antigen from CHO cells.
Minimal lot-to-lot variation • Hydrophilic backbone gives low non-specific interactions.
Low risk of denaturation of relatively strong hydrophobic solutes.

Maximum operating pressure

Base matrix: 250-400 cm/h, 100 kPa, XK 50/60 column, bed height 25 cm

Sample preparation

The sample should be dissolved in start buffer. Alternatively the sample may be transferred to start buffer by dialysis or by buffer exchange using a HiTrap Desalting or a PD-10 Desalting columns. The viscosity of the sample should not exceed that of the buffer. For normal aqueous buffer systems, this corresponds to a protein concentration of approximately 50 mg/ml.

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Butyl-S Sepharose 6 Fast Flow

Matrix
6% cross-linked agarose
Particle Size
45 µm-165 µm
Average particle size
~90 µm
Ligand
Butyl-S
Ligand density
Approx 10 µmol Butyl-S/ml medium
Recommended flow rate
Approx. 400 cm/h at 1 bar (100 kPa, 14.5 psi), XK 50/30 column, bed height 15 cm.
Recommended column height
25 cm
Chemical stability
Stable in commonly used aqueous buffers: 1 mM HCl, 1 M NaOH, 30% isopropanol, 50% ethylene glycol, 70% ethanol, 6 M guanidinehydrochloride, 8 M urea.
pH working range
3–13
CIP stability
2–14
Temperature stability
4°C to 40°C
Autoclavable

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20 min at 121°C in 0.05 M sodium phosphate pH 7, 1 cycle.

Storage

4 to 30°C, 20% Ethanol

Shipping

20% ethanol

Evaluation of Packing

The best method of expressing the efficiency of a packed column is in terms of the height equivalent to a theoretical plate (HETP) and the asymmetry factor (As). The values are easily determined by applying a test sample such as 1% acetone solution or sodium chloride to the column.

Binding

Those salts which cause salting-out (e.g., ammonium sulphate) also promote binding to hydrophobic ligands. The column is equilibrated and the sample is applied in a solution of high ionic strength. A typical starting buffer is 1.7 M (NH4)2SO4 , which is just below the concentration employed for salting-out proteins. Hydrophobic interactions are weaker at lower temperatures. This must be taken into account if chromatography is done in a cold room.

Equilibration

To equilibrate, pump approximately 100 mL of start buffer through the column at a flow rate of 3.5 mL/min. The column is fully equilibrated when the pH and/or conductivity of the effluent is the same as the start buffer.

Elution

Reducing the concentration of salting-out ions in the buffer with a decreasing salt gradient.
Increasing the concentration of chaotropic ions in the buffer with an increasing gradient.
Eluting with a polarity-reducing organic solvent added to the buffer.
Eluting with detergent added to the buffer.

Cleaning-in-place

Remove precipitated proteins: Wash the column with 4 bed volumes of 0.5–1.0 M NaOH solution at 40 cm/h, followed by 2–3 bed volumes of water.

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CREATIVE BIOMART®
Chromatography

Butyl-S Sepharose 6 Fast Flow

Remove tightly bound hydrophobic proteins, lipoproteins, and lipids: Wash the column with 4–10 bed volumes of up to 70% ethanol or 30% isopropanol. (Apply gradients to avoid the formation of air bubbles when using high concentrations of organic solvents.) Alternatively, wash the column with detergent in a basic or acidic solution, for example 0.5% non-ionic detergent in 1 M acetic acid. Wash at a flow rate of 40 cm/h. Residual detergent can be removed by washing with 5 bed volumes of 70% ethanol.

Sanitization
Sanitization is the use of chemical agents to inactivate microbial contaminants. Sodium hydroxide (NaOH) is a commonly used sanitizing agent. A concentration of 0.5–1.0 M NaOH with a contact time of 30–60 min is effective for most microbial.
Pack size
200 mL
BioProcess resin
Yes

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