

HiTrap rProtein A FF

Product Information

Cat#No# Hi-022P

Product Overview

HiTrap rProtein A FF columns are prepacked with rProtein A Sepharose Fast Flow for purification and fractionation of IgG subclasses and IgG from different sources, including cell supernatants and ascites.

Characteristic

Quick, convenient preparative purification of monoclonal antibodies.

Well-established protein A-based resin used in many approved MAb processes.

Recombinant protein A exhibits similar Fc region specificity to that of native protein A but shows enhanced binding capacity and fewer regulatory concerns due to the total absence of mammalian culture in the ligand production and purification.

Simple operations with a syringe, pump, or high-performance chromatography system, such as ÄKTA design.

Maximum operating pressure

5 bar [0.5 MPa] (70 psi)

Sample preparation

The sample should be adjusted to the composition of the binding buffer. This can be done by either diluting the sample with binding buffer or by buffer exchange using HiTrap Desalting, HiPrep 26/10 Desalting or PD-10 Desalting columns. The sample should be filtered through a 0.45 µm filter or centrifuged immediately before it is applied to the column. Never apply a turbid solution to the column.

Matrix

cross-linked agarose, 4%, spherical

Average particle size

~ 90 µm

Ligand

Recombinant protein A, (E. coli)

Dynamic binding capacity

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~ 35 mg human IgG/mL resin

Recommended flow rate

< 4 ml/min

Recommended column height

25 mm

Chemical stability

Stable to commonly used aqueous buffers, 6 M guanidine hydrochloride, 2% benzyl alcohol, 1 mM NaOH (pH 11), 0.1 M sodium citrate/HCl (pH3), 20% ethanol.

pH working range

3 to 10

CIP stability

3 to 12

Temperature stability

2°C to 40°C

Storage

4 to 30°C, 20% Ethanol

Binding buffer

20 mM sodium phosphate, pH 7.0.

Elution buffer

0.1 M sodium citrate, pH 3 to 6.

Purification procedures

1. Prepare collection tubes by adding 60 to 200 µL of 1 M Tris-HCl, pH 9.0 per mL of fraction to be collected.
2. Remove the stopper from the inlet and the snap-off end at the column outlet.
3. Connect the column to the system with 1/16" male connectors.
- 4 Wash out the ethanol preservative with at least 5 column volumes of distilled water or binding buffer.

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5. Regenerate the column with 5 column volumes of elution buffer.
6. Equilibrate the column with 5 to 10 column volumes of binding buffer.
7. Apply the sample, using a syringe fitted to the luer connector or by pumping it onto the column.
8. Wash with 5 to 10 column volumes of binding buffer or until no material appears in the effluent. Excessive washing should be avoided if the interaction between the protein of interest and the ligand is weak, since this might decrease the yield.
9. Elute with elution buffer. 2 to 5 column volumes is usually sufficient, but other volumes (or different elution buffer) will be required if the interaction is difficult to break.
10. The purified IgG fractions can be buffer exchanged using HiTrap Desalting, HiPrep 26/10 Desalting or PD-10 Desalting columns if necessary.

Pack size

5 × 1 mL

Dimensions

7 × 25 mm

Column volume

1 mL

Column i.d.

7 mm

Column hardware pressure limit

5 bar (0.5 MPa)
