

nProtein A Sepharose Fast Flow antibody purification resin

Product Information

Cat#No# nP-338C

Product Overview

nProtein A Sepharose 4 Fast Flow is a native protein A resin for purification of monoclonal and polyclonal antibodies.

For the purification of monoclonal and polyclonal antibodies at both laboratory and process scale.

Used in routine commercial polyclonal and monoclonal antibody purification and production.

Free from animal-derived components.

Well suited to immunoprecipitation procedures.

Hydrophilic base matrix ensures low levels of nonspecific binding and low levels of host cell-derived impurities in the elution pool. Fulfills industrial demands for security of supply, robust performance, and regulatory support.

Description

nProtein A Sepharose 4 Fast Flow is native protein A coupled to Sepharose 4 Fast Flow. It has nearly twice the total IgG binding capacity of Protein A Sepharose CL-4B, and is an excellent adsorbent for recovery and purification of monoclonal antibodies from cell culture at both laboratory and process scale.

nProtein A Sepharose 4 Fast Flow has been developed and tested in cooperation with leading manufacturers of purified monoclonal antibody products, and is used in routine commercial production.

Characteristic

Low leakage of protein A.

Used in large-scale FDA-approved processes.

Manufactured without using animal-derived components.

Applications

The most important application area for nProtein A Sepharose 4 Fast Flow is the purification of monoclonal antibodies from cell culture. High IgG capacity and high flow velocities make the resin ideal for both laboratory- and process-scale separations.

There is a natural diversity between the different subclasses of IgG and even within subclasses. Therefore the binding and elution system must be optimized for every monoclonal antibody to be purified.

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Maximum operating pressure

< 0.1 MPa in a XK 50/60 column with 5 cm diameter and 25 cm bed height (at 20°C using buffers with the same viscosity as water).

Ligand Coupling Method

Cyanogen bromide activation

Matrix

cross-linked agarose, 4%, spherical

Average particle size

~90 µm

Ligand density

6 mg Protein A/ml medium

Dynamic binding capacity

>35 mg human IgG/mL

Recommended flow rate

30 to 200 cm/h

Recommended column height

25 cm

Chemical stability

Stable in aqueous buffers commonly used in Protein A chromatography, 6 M guanidine-HCl, 70% ethanol, 3 M NaSCN, 0.1 M glycine (pH 3.0), 2% benzyl alcohol or 20 % ethanol.

pH working range

3–9

CIP stability

2–10

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Temperature stability

4°C to 40°C

Storage

2 to 8°C, 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 (A_s). These values are easily determined by applying a sample such as 1% acetone solution to the column. Sodium chloride can also be used as a test substance. Use a concentration of 0.8 M NaCl in water as sample and 0.4 M NaCl in water as eluent.

Equilibration

After packing, and before a chromatographic run, equilibrate with working buffer by washing with at least 5 bed volumes.

Regeneration

After each separation cycle, regenerate the resin bed by washing with approximately three column volumes of 0.1 M citrate buffer, pH 3 until the baseline is stable.

Cleaning-in-place

As cleaning protocol, 6 M guanidine hydrochloride can be used. Phosphoric acid (100 mM) has also been used for cleaning. To remove hydrophobically-bound substances, a solution of non-ionic detergent or ethanol is recommended.

Sanitization

Wash the packed column with 2% hibitane/20% ethanol or 70% ethanol.

Scaling up

1. First consider the residence time. This should not be shorter than the time established in the small scale experiments.
2. Select bed volume according to required binding capacity.
3. Select column diameter to obtain a bed height of 5 to 30 cm so that high flow velocities can be used. Max.

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flow velocity is approx. inversely proportional to the bed height. Expect to operate at no more than 70% of the max. flow velocity.

4.Keep sample concentration and the ratio of gradient volume/resin volume constant.

Pack size

5 mL

Maximum flow velocity

200 cm/h

Dimensions

5 cm
