

HiTrap CM Sepharose FF

Product Information

Cat#No# Hi-226P

Product Overview

HiTrap CM FF is prepacked with CM Sepharose Fast Flow, and is a weak cation exchanger for small-scale protein purifications as well as screening of binding and elutions conditions.

Description

Q, SP, DEAE and CM Sepharose Fast Flow are based on a robust, 6% highly cross-linked beaded agarose matrix with good flow properties and high loading capacities. ANX Sepharose 4 Fast Flow (high sub) is based on 4% highly cross-linked beaded agarose. This results in a resin with higher porosity, which is particularly useful for the purification of high molecular mass proteins.

Characteristic

Convenient and affordable for fast, easy ion exchange separations either alone or connected in series.
The industry standard for ion exchange chromatography.
High flow rates and good scale-up potential.
Use a weak ion exchanger if the selectivity of a strong ion exchanger is insufficient.
Predictable scale-up.

Applications

For method scouting, group separations, sample concentration and sample clean-up of charged biomolecules.

Maximum operating pressure

5 bar [0.5 MPa] (70 psi)

Sample preparation

The sample must be adjusted to the composition of the start buffer by buffer exchange using HiTrap Desalting, HiPrep 26/10 Desalting or PD-10 columns. The sample must be filtered through a 0.45 µm filter or centrifuged immediately before it is applied to the column.

Metal ion capacity

HiTrap CM Sepharose FF

0.09–0.13 mmol H⁺/ mL resin

Matrix

Cross-linked agarose, 6%, spherical

Ionic Exchanger Type

Weak cation

Particle Size

45–165 µm

Average particle size

~ 90 µm

Dynamic binding capacity

~ 50 mg Ribonuclease A/ mL resin.

Recommended flow rate

< 4 ml/min

Recommended column height

25 mm

Chemical stability

Stable to commonly used aqueous buffers, 1.0 M NaOH, 8 M urea, 6 M guanidine hydrochloride, 70% ethanol.

pH working range

4 to 13

CIP stability

2 to 14

Storage

4 to 30°C, 20% Ethanol

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Purification procedures

1. Fill the syringe or pump tubing with start buffer (low ionic strength). 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.
 2. Remove the snap-off end at the column outlet.
 3. Wash out the preservatives with 5 column volumes of start buffer, at 1 mL/min for HiTrap 1 mL and 5 mL/min for HiTrap 5 mL.
 4. Wash with 5 column volumes of elution buffer (start buffer with 1 M NaCl).
 5. Finally equilibrate with 5 to 10 column volumes of start buffer.
 6. Apply the sample at 1 mL/min for HiTrap 1 mL and 5 mL/min for HiTrap 5 mL using a syringe fitted to the luer connector or by pumping it onto the column.
 7. Wash with at least 5 column volumes of start buffer or until no material appears in the eluate.
 8. Elute with 5 to 10 column volumes of elution buffer, see "Choice of gradient type".
 9. The purified eluted fractions can be desalted using a HiTrap Desalting, HiPrep 26/10 Desalting or a PD-10 column if necessary.
 10. After completed elution, regenerate the column by washing with 5 column volumes of regeneration buffer (start buffer with 1 M NaCl) followed by 5 to 10 columns volumes of start buffer. The column is now ready for a new sample.
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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)

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Functional group

-O-CH₂COO
