Smac Q

Index

1. Product Description.....1
2. Purification Procedure.....1
3. Cleaning-in-Place.....2
4. Troubleshooting.....3
5. Related Products.....3

1. Product Description

Ion exchange resins are widely used in biomedical and bioengineering for separation and purification of proteins, nucleic acids and polypeptides.**Smac Q** based on a highly rigid agarose base matrix that offers outstanding pressure/flow properties, optimized pore structure,and very high chemical stability to support CIP procedures.

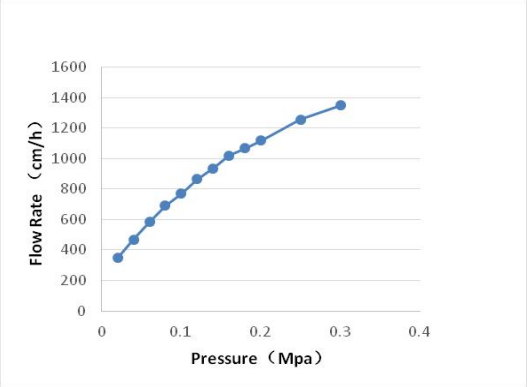


Fig.1. A typical pressure/flow rate curve

Smac Q is a strong anion exchange resin. The ion exchange group is a quaternary amine group.see below.

$-O-CH_2CHOHCH_2OCH_2CHOHCH_2N^+(CH_3)_3$

Table 1. Characteristics of **Smac Q**

Item	Description
Matrix	Highly cross-linked agarose
Ion exchange type	Strong anion
Ion exchange capacity	0.16-0.23 mmol Cl^-/ml medium
Particle Size	45-165 μm
Flow rate	< 700cm/h
pH stability	2-12
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.

2.2 Sample Preparation

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

2.3 Packing Columns

1) Remove air from the column dead spaces by flushing the end-piece and adapter with packing buffer. Make sure no air has been trapped under the column net.

2) Close the column outlet leaving the net covered with packing buffer.

3) Resuspend the beads stored in its container by shaking (avoid magnetic stirring the sedimented medium). Pouring the slurry down a glass rod held against the column wall will minimize the introduction of air bubbles.

If using a packing reservoir, immediately fill the remainder of the column and reservoir with packing buffer. Mount the adapter or lid of the packing reservoir and connect the column to a pump. Avoid trapping air bubbles under the adapter or in the inlet tubing.

4) Open the bottom outlet of the column and set the pump to run at the desired flow velocity. Ideally, the medium is packed at a constant pressure of approximately 3 bar (0.3 MPa). If the packing equipment does not include a pressure gauge, use a packing flow velocity of Approximately 200 cm/h (10 cm bed height, 25°C, low viscosity buffer). If the recommended pressure or flow velocity can not be obtained, use the maximum flow velocity the of the pump. This should also give a well-packed bed. Do not exceed 75% of the packing flow velocity in subsequent chromatographic procedures.

5) When the bed has stabilized, mark the position of the bed surface. Close the bottom outlet and stop the pump.

If using a packing reservoir, disconnect the reservoir and fit the adapter to the column. If using the column, carefully place the top filter on top of the bed before fitting the adapter.

6) With the adapter inlet disconnected, adjust the adapter to the bed surface.

7) Connect the pump, open the bottom outlet and continue packing. The bed will be further compressed at this point and a space will be formed between the bed surface and the adapter.

8) Close the bottom outlet. Disconnect the column inlet and push the adapter a further 2-3mm. Connect the pump. The column is now ready to use.

2.4 Sample Purification

1) Fill the pump tubing with binding buffer. Connect the column to purification system, “drop to drop” to avoid introducing air into the column.

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.

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.

2.5 Analysis

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

3. Clean-in-Place

After each separation, elute any reversibly bound material either with a high ionic strength solution (e.g. 1M NaCl in buffer) or by increasing pH. Regenerate the media by washing with at least 5 bed volumes of buffer, or until the column effluent shows stable conductivity and pH values.

Cleaning-in-place (CIP) is a cleaning procedure that removes contaminants such as lipids, precipitates, or denatured proteins that may remain in the packed column after regeneration. Regular CIP also prevents the build-up of these contaminants in the media bed and helps to maintain the capacity, flow properties and general performance of the media.

A specific CIP protocol should be designed for each process according to the type of contaminants present. CIP cycle is generally recommended every 1-5 separation cycles.

Remove the ionically bound proteins

Wash with 3-4 column volumes of 2M NaCl. Contact time 10-15min.

Remove the precipitation or hydrophobically bound proteins or lipoproteins

Wash with at least 2 column volumes of 1M NaOH . Contact time 1-2h.

Remove lipids and very hydrophobic proteins

Wash with 2-4 column volumes of 0.5% non-ionic detergent,70% ethanol or 30% isopropanol. Contact time 1-2h.

4. Troubleshooting

Problem	Probable Cause	Solution
Back pressure is too high	Column is clogged	Cleaning in place(part 3).
	Sample solution contains precipitate	Filtering the sample solution by passing them through a 0.22μm or 0.45μm filter.
Eluate is not pure	The medium repeat too much times.	Cleaning in place(part 3).
	Wash is not enough.	Increase the volume of Wash Buffer.

5. Related Products

Product	Cat. No.	Size
Smac Q	SI019025	25 ml
	SI019100	100 ml
	SI019500	500 ml
	SI01901L	1 L
	SI01910L	10 L
IexCap Smac Q	SI019C11	1X1 ml
	SI019C51	5X1 ml
	SI019C15	1X5 ml
	SI019C55	5X5 ml