



Sartobind® Capsules for Capture Chromatography

Ion Exchange Chromatography with Sartobind® Membrane Adsorbers



Void volume optimization for cylindrical scale down capsules

Sartobind nano 3 ml is the scale down model for the Sartobind Jumbo 5 l ion exchange membrane adsorber. It contains void volume optimized flow channels which differentiates this unit from adsorbers developed mainly for flow through applications.

Introduction

Sartobind membrane adsorbers with 8 mm bed height (30 membrane layers) can be used for flow through and bind and elute (B&E) application as flow channels have been optimized for smallest void volume. Such optimization is essential to achieve sharp breakthrough curves, small elution volumes below two bed volumes (column volumes) and no back mixing of already separated solute samples.

The capsule design keeps the advantages of disposable Sartobind SingleSep® capsules with 4 mm bed height. They can be used for flow through applications due to the high flow rate, the clear scale and usage of same materials through the product line.

On the other hand the 8 mm capsules provide more membrane per capsule for higher binding capacity, they are reusable and meet the expectations for small elution volumes and peak resolution as shown here.

Fig. 1: Top view of Sartobind nano 3 ml (8 mm)



Void volume optimization in 5 l scale: Sartobind Jumbo capsule

The Sartobind Jumbo 5 l contains equivalent optimization details and an additional central core to reduce void volume.

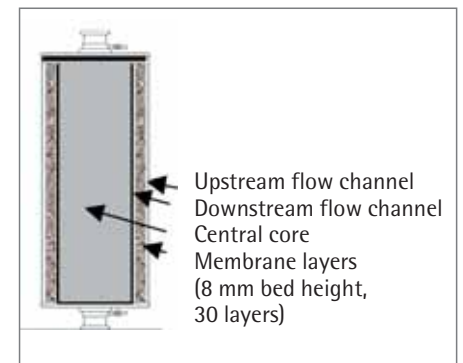


Fig. 2: Section view of Sartobind Jumbo 5 l

Scalability of Sartobind Q nano 3 ml to Q Jumbo 5 l

Bovine serum albumin (BSA) 2 g/l in 10 mM potassium phosphate pH 7.4 was loaded at a flow rate of 4 bed volumes (BV) per minute on three different lots of Sartobind Q nano 3 ml and Q Jumbo 5 l. The breakthrough curves were normalized to bed volume for comparison. The six curves show equivalent shape of breakthrough behaviour as well as elution.

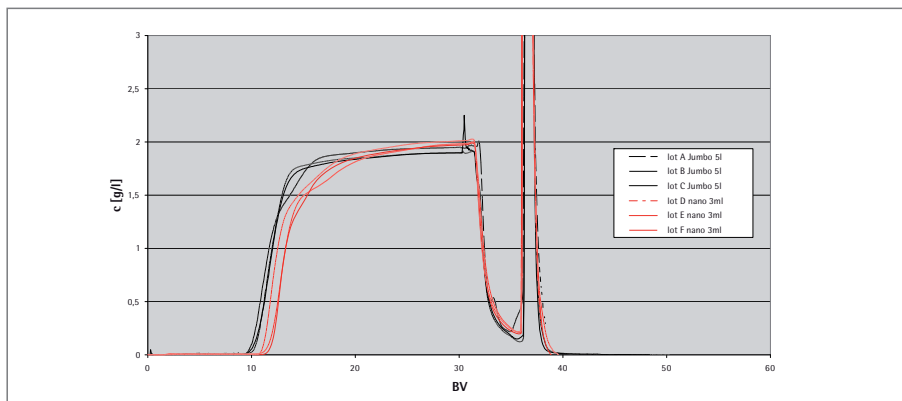


Fig. 3: Breakthrough curves of Sartobind Q Jumbo 5 ml and Q nano 3 ml normalized to the bed volume

Analysis of elution volume

For the analysis a 1 % acetone in 10 mM potassium phosphate buffer pH 7.4 was ran over each three different nano and Jumbo capsules (Fig. 4). At 50 % acetone breakthrough, the breakthrough curve indicates a volume of ~ 1.6 bed volumes. Such elution volumes are similar to conventional void volume optimized columns.

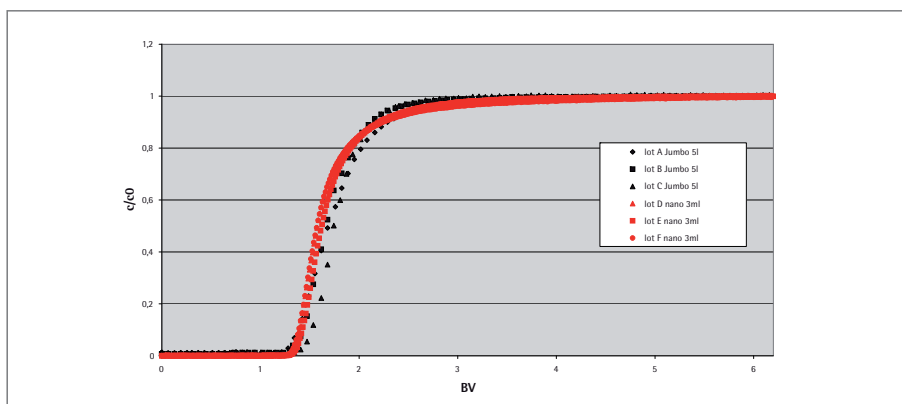


Fig. 4: Breakthrough of acetone in Sartobind Jumbo 5 l and nano 3 ml normalized to the bed volume

Bind & Elute performance of Sartobind Q Jumbo

Sartobind Q Jumbo was loaded with 2 g/l BSA in 10 mM potassium phosphate buffer pH 7.4 at a flow rate of 4 BV per minute (Fig. 5). The complete cycle of loading, washing and eluting with 220 l total buffer volume was achieved within 11 minutes. The profile shows a sharp breakthrough with a small elution volume of about 2 BV (10 l) and a dynamic binding capacity of ~ 90 g.

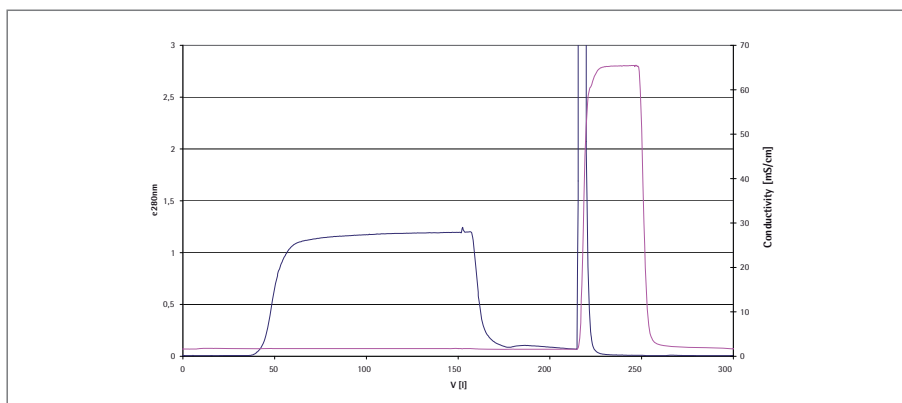


Fig. 5: Breakthrough curve of Sartobind Q Jumbo 5 l

Separation of myoglobin, cytochrome c and lysozyme with Sartobind S nano 3 ml

Sartobind nano 3 ml can be used for analytical protein purifications. The flow pattern display good resolution comparable to chromatographic column technology (Fig. 6). The main difference to columns is the high speed at which purification can be achieved. Flow rate was 10 ml/min (3.3 BV/min)

Pre-conditioning	2 M NaCl in equilibration buffer	20 BV
Equilibration	20 mM Tris pH 7.4, 1.8 mS/cm	25 BV
Loading of protein mixture	~ 1-1.5 mg/ml for protein 1, 2 and 3	500 µl
Wash	20 mM Tris pH 7.4, 1.8 mS/cm	4 BV
Elution by linear gradient	2 M NaCl in equilibration buffer	16 BV

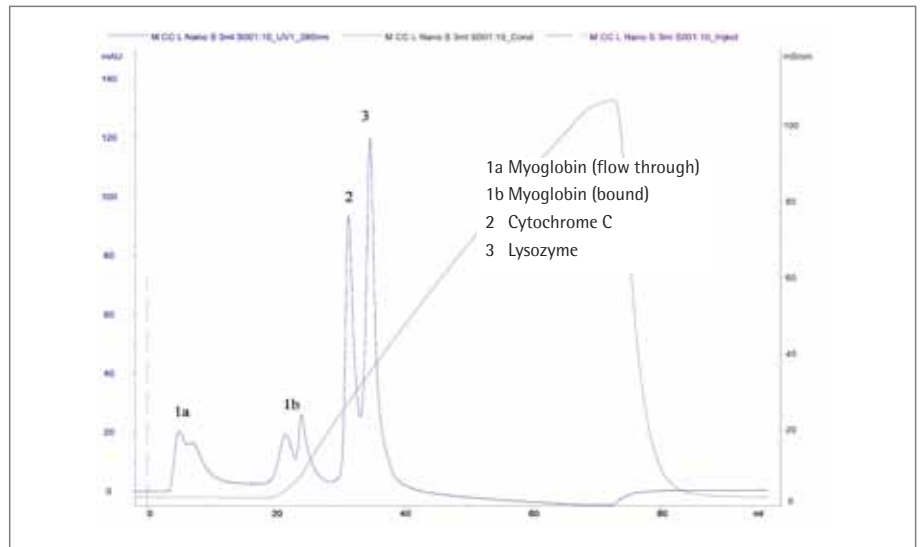


Fig. 6: Separation of myoglobin, cytochrome C and lysozyme with Sartobind S nano 3 ml

Separation of alpha-chymotrypsinogen A, ribonuclease and lysozyme with Sartobind S nano 3 ml

The second example (Fig. 7) displays as well high resolution of three sample proteins within 10 minutes.

Pre-conditioning	2 M NaCl in equilibration buffer	20 BV
Equilibration	20 mM NaAC pH 5.0, 1.1 mS/cm	25 BV
Loading of protein mixture	~ 1-1.5 mg/ml for protein 1, 2 and 3	500 µl
Wash	20 mM NaAc pH 5.0, 1.1 mS/cm	4 BV
Elution by linear gradient	2 M NaCl in equilibration buffer	100 BV

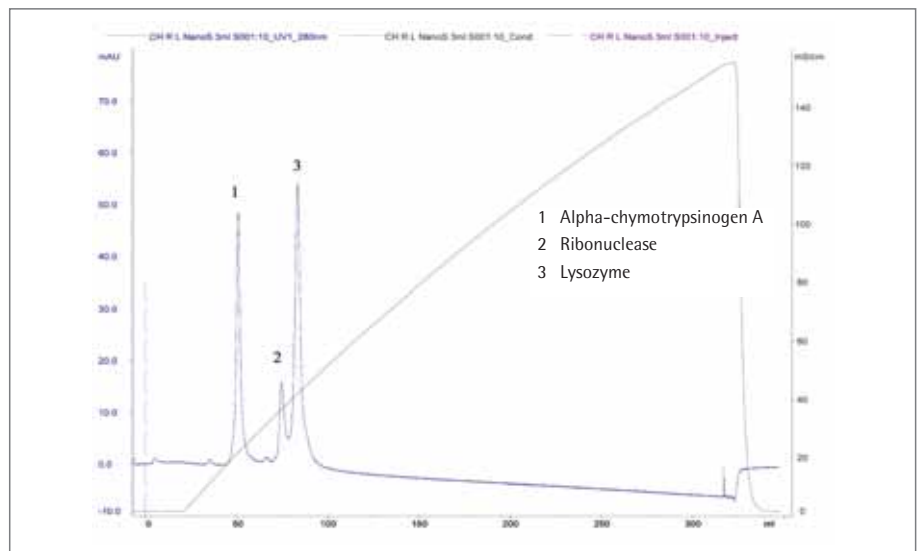


Fig. 7: Separation of alpha-chymotrypsinogen A, ribonuclease and lysozyme with Sartobind S nano 3 ml

Summary

Sartobind Jumbo and its void volume optimized scale down unit, Sartobind nano 3 ml, can be used for bind and elute applications. They show comparable and scaleable break-through behavior and elution volumes.

Sartobind Membrane Adsorbers with 8 mm bed height are versatile as they can be used in flow through and bind and elute separations.

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