Simplifying Progress

Intensification of mAb Processes: Leveraging Sartobind® Rapid A and a Fully Connected Membrane-Based DSP

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Introduction

In the field of monoclonal antibody (mAb) purification, there is an increasing demand for high-performance chromatography membranes that are ready-to-use and support a "one-batch-one device" manufacturing strategy. The new Protein A capture technology Sartobind® Rapid A increases productivity 10-fold compared to traditional resins (203 g/L vs. 14 g/L, respectively) when used in rapid cycling conditions. The membranes also show similar performances for dynamic binding capacity (DBC), yield, and host cell protein (HCP) | host cell DNA (hcDNA) removal. As such, the Sartobind® Rapid A supports a new generation of fully membrane-based purification platforms.

Protein A › IEX › Polishing

The first milestone in a fully membrane-based process is implementing a competitive double-flowthrough polishing process with connected Sartobind® Q and Sartobind® S. Comparable purity and yield were obtained (> 98% for each flowthrough step) with a strong footprint reduction of the purification process. The second step to a full membrane process is combining the Resolute® MCC multi-column technology with Protein A and anion exchange (AEX) and cation exchange (CEX) membranes in parallel batch mode. This drives further productivity increases (> 400 g/L/h) compared to a resin-based multi-column chromatography process (< 200 g/L/h).

This innovative Sartobind® Rapid A combined with process intensification solutions demonstrates that alternative mAb purification platforms are a highly competitive alternative to classic resin-based approaches.

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Convecdiff Membrane Sartobind® Rapid A vs Purely Convective | Diffusive Materials

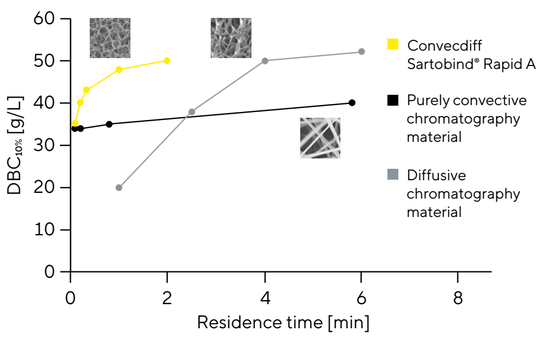
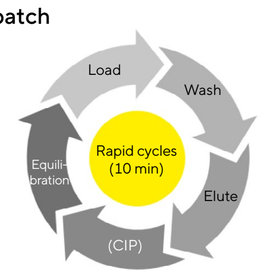


Figure 1: *$SDBC_{10\%}$ As a Function of Residence Time for Commercially Available Materials and the Convecdiff Sartobind® Rapid A*

Convecdiff materials support high DBC and flowrate to facilitate short cycle time

Sartobind® Rapid A enables lifetime capacity utilization in one single-batch

- Fast cycles: 10 – 15 min
- ~30 – 150 cycles/batch



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Comparability of Sartobind® Rapid A and Standard Resin[1]

We compared critical process paramters (CPPs) and critical quality attributes (CQAs) between Sartobind® Rapid A and standard Protein A resins. Both materials were tested with the same feed material. The analyzed data show a very good comparability of Sartobind® Rapid A with the Protein A resin. The membrane showed superior performance in DNA reduction and Protein A leaching, with a 14.5-fold increase in productivity.

	Sartobind® Rapid A	Protein A Resin
DBC10% [g/L]	42.9 ± 0.8	30.4 ± 0.5
Residence time [min]	0.2	4.0
Yield [%]	94.7 ± 0.2	96.4 ± 0.4
HCP reduction [LRV]	2.2 ± 0.2	2.3 ± 0.1
hcDNA reduction [LRV]	2.9 ± 0.2	2.3 ± 0.1
Protein A leached [ppm]	2.7 ± 0.7	6.7 ± 0.3
Av. Productivity [g/L*h]	203.6	14.1

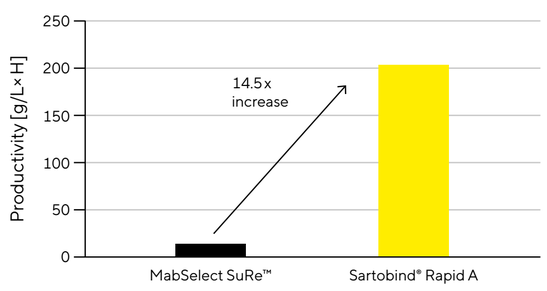

Figure 2: *Productivity Comparison of Standard Protein A Resin to Sartobind® Rapid A*

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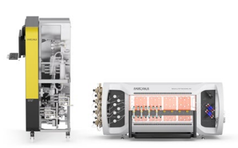
Levels of Intensification for Downstream Processing

Level 0	Level 1	Level 2	Level 3	Level 3.1
Standard batch, standalone UO	**Intensified, standalone UO:** increases the individual step productivity; higher cycling (RCC or MCC), improved buffer management (ILD, ILC), high DBC resins; pooling tanks	**Connected process:** at least 2 UO; subsequent steps started before first step is finished; could be staggered batch; may have pooling tanks; **software orchestration** is beneficial; might be called clustered or linked process	**Continuous process:** more integrated case of a connected process; **steady state flow;** only small intermediate (surge) tanks; software orchestration is a must-have; **long run times; closed processing**	**Flow-through continuous process:** further integrated case with complete steady state flow. **All bind and elute steps are replaced with flow through mode.** Molecule does not stop – ideally no intermediate (surge) tanks



Resolute® RCC MU

Resolute® BioSMB PD

Resolute® BioSC Pilot

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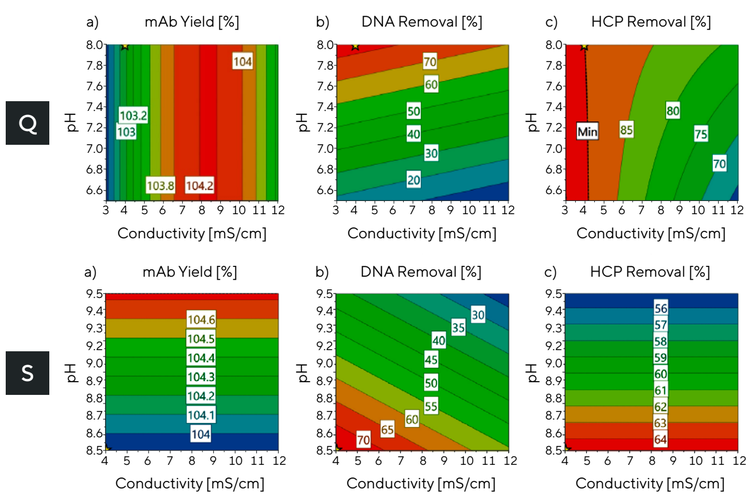
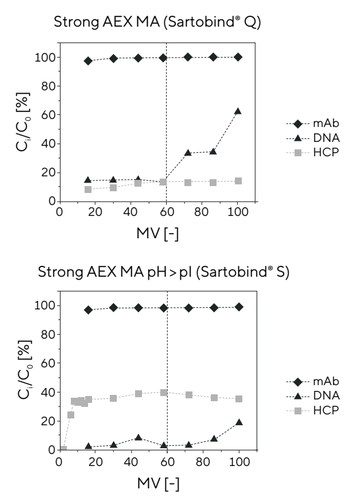
Double Flow-Through With Membrane Adsorbers[2]

Step 1: Design of Experiments (DoE) to Define Buffer Conditions



Step 2: Breakthrough Curves



Note. Sartobind® Rapid A - Beta Test Opportunity. DoE done with MODDE® 13

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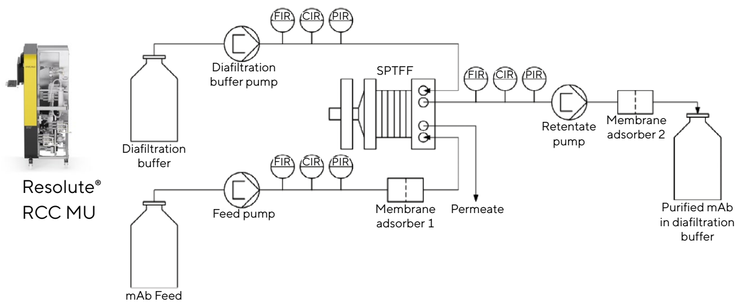
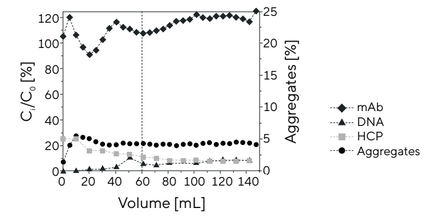
Chromatography Membrane—Towards a Connected Process[2]

Step 3: Connected Processing



- Yield > 98% (per step)
- Productivity: 240 $g_{(mAb)} \times L^{-1}_{(membrane)} \times h^{-1}$
- DNA: 2 ppm
- HCP: 29 ppm



- Comparable purity with classical process
- No column packing
- Rapid cycling chromatography (RCC) enables strong footprint reduction
- Full single batch use

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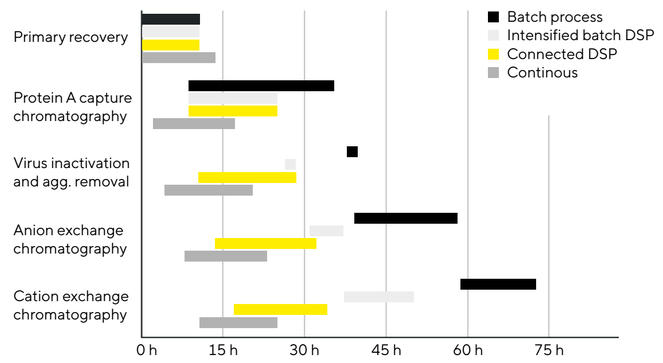
Downstream Intensification Reduces Processing Times

- Each step starts before the previous one ends
- This enables processing the sub-batches from Protein A elution
- Reduction of intermediate tanks and column size
- Lower footprint
- Lower OPEX



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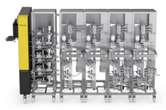
The Power of Connected Membrane Processes

Cascade	Resin (MCC Connected Process)	Sartobind®
Number of steps	3	3
Final yield [%]	83	86
DNA [ppb]	<3	<3
HCP [ppm]	3	30
HMW [%]	0.2	0.3
Total process time [h]	10.4	4.0
Av. Productivity [g/h]	179	465

- Connecting the process and using MCC or parallel batch multiplies productivity or reduces costs
- Comparable purity and yield
- Lower footprint



Resolute® BioSC Pilot

Clarified harvest

Sartobind® Rapid A (9.6 L @ 45 g/L)
3 × 3.2 L membrane

Sartobind® Q (1.6 L @ 500 g/L)

Sartobind® S (1.6 L @ 500 g/L)

Conclusion

Due to their inherent structural characteristics, Sartobind® Rapid A membranes offer unique possibilities for fully membrane-based ultra-fast mAb purification processes

- Sartobind® membranes are ready-to-use and enable a one-batch, one-device manufacturing strategy
- Highly competitive DBC
- Short cycle time (< 30 min)
- High number of cycles per batch (up to 150)
- Double flow-through polishing with Sartobind® Q | S
- Full membrane process with ultra-high productivity (3x compared to connected resin-based process)
- The innovative agarose platform is scalable and robust for a wide variety of mAbs with limited back-pressure at large scale
- Availability of modular cassette format enables scaling to large production processes

Abbreviations:
ASAP: Accelerated Seamless Antibodies Purification, EASY: Full flow-through process, ILC: Inline Conditioning, ILD: Inline dilution, MoBiDiK: Modular Bioproduction, disposable and continuous, MCC: Multi-column chromatography, RCC: Rapid cycling chromatography, UO: Unit operation

1 Grünberg et al. (2022). Membranes.
2 F. Schmitz et al. (2023). Integrated double flow-through purification of monoclonal antibodies using membrane adsorbers and single-pass TFF. Biochemical Engineering Journal 195.