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Catalent
BIOLOGICS



Multi-column chromatography implementation and scale-up

Your Challenge

- ▶ You deal with increasingly challenging antibody purification throughput bottlenecks and need to increase the efficiency of your capture process.
- ▶ You need a straightforward proof of concept showing implementation and scale up of a multi-column chromatography process.

Our Solution

Octave and SkillPak Multi-Column Chromatography Platform

- ▶ Scalable systems, software, and columns

What was done?

- ▶ An existing purification process was transferred to a multi-column chromatography format at a 100 g pilot scale and scaled up to a 1 kg clinical scale.

What was the result?

- ▶ Resin volume and buffer savings were achieved by transferring to MCC, while repeatable impurity profiles were achieved with the scaled-up process.

Existing antibody purification processes can be transferred to a multi-column format and scaled up with the Octave BIO process development system and the Octave PRO GMP ready system. Catalent now considers MCC as an option of their process intensification offering alongside their GPEX® Lightning cell line development technology.

Your Benefit

Observe the measurable benefits of intensified purification with multi-column chromatography at a clinical scale.

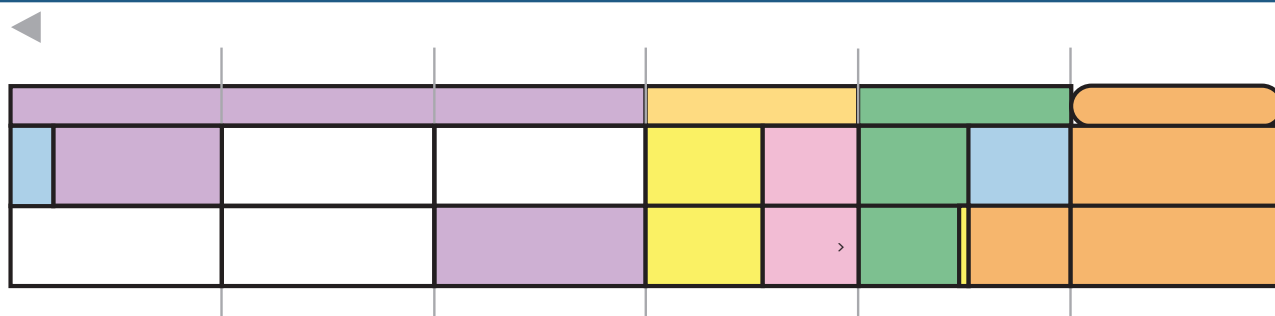


Intensified purification of antibody therapies

TOSOH BIOSCIENCE

<https://www.separations.eu.tosohbioscience.com/products/chromatography-instruments/multi-column-chromatography/octave-pro-system>

➤ **Figure 1.** Process flow diagram of the 6-column scale up MCC configuration.



In this configuration there are three columns in the capture (Feed) zone (columns 1–3), one column in the Wash 2 zone (columns 6), and one column each in the W3 & Elute and CIP & Equilibrate zones (columns 5 and 4, respectively). Each process step has a dedicated pump and inlet and a defined outlet. During regular time intervals programmed in PROComposer, specified inlet and outlet ports are switched to the next process step via a series of valves arrayed in a compact three-dimensional valve block (e.g. Octave PRO System) to produce a continuous cycle.

Utilizing the same MethodWizard and PROComposer software, a six-column method (*Figure 1*) for increased productivity, was designed utilizing the same TOYOPEARL AF-rProtein A HC-650F resin, obtained in a prepacked SkillPak PRO format (14 × 5.1 cm). Because of the similarity between the process design, monitoring, and control software, the small-scale (Octave BIO) and large-scale (Octave PRO), process scale-up was straightforward.

The process parameters of the scaled-up experiments are summarized in *Table 3*. The two runs presented here were executed with the designed process, each processing a 250L bioreactor at similar expression levels (5.6 and 6.2 g/L) for three steady state cycles. After Protein A capture, the eluate was neutralized to pH 6.8 - 7.2, and the product impurity profile (aggregation, HCP, DNA) and quality (monomer %) were assessed through offline analytical testing.

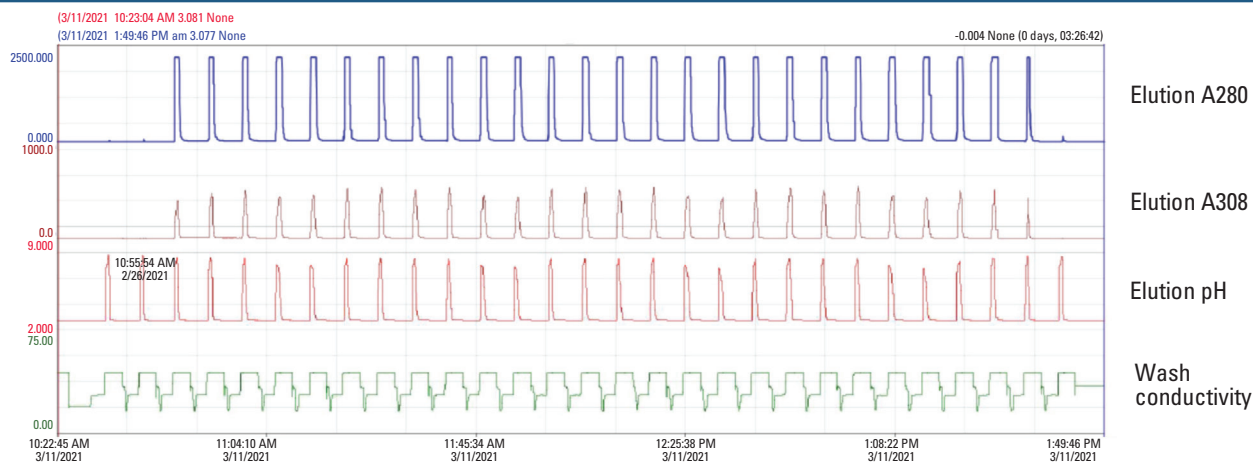
➤ **Table 3.** Key process parameters for the scaled-up multi-column processes.

Process Parameters	Run 1	Run 2
Column Size	14 × 5.1 cm, 0.785 L	
Number of Columns	6	
Total resin volume	4.71 L	
Residence time	0.5 min	
Expression Levels	5.6 g/L	6.2 g/L
Total Feed Loaded	206.4 L, 1123.4 g	185.3 L, 1148.6 g
Process time	3.4 h	3.1 h

➤ **Figure 2.** Operation of the Octave PRO at Catalent facilities.

➤ **Figure 3.** The SkillPak PRO columns installed for the scaled-up MCC process.

Figure 4. Process monitoring sensor profiles as viewed on the Octave PRO.



Results

The Octave PRO, along with SkillPak PRO columns, shows consistent column performance during the execution of the kilogram scale MCC process. Qualitatively, the sensor profiles at steady state for Elution UV, Elution pH, and Wash conductivity are consistent over four cycles (Figure 4).

The measurable results of the two runs are summarized in Table 4. The offline analysis of product quality confirms the consistent performance of the MCC purification process. Aggregate impurities were at or below 0.5% for both runs, while host cell protein and DNA levels were reduced consistently by about 1.5 logs for both runs, showing replicable impurity clearance. Product monomer levels remained consistently around 95% as well.

Table 4. Key process parameters for the scaled-up multi-column processes.

Scale Up Results	Run 1	Run 2
Yield	1,041.5 g, 93%	1,055.5 g, 92%
Steady State Productivity	76 g/L/hr	83 g/L/hr
% aggregate	0.5%	<0.5%
% monomer	95%	94%
HCP log reduction	1.59	1.55
DNA log reduction	1.49	1.44

Regarding process productivity, both processes maintained the efficiency gains of transferring a batch process to MCC and even exceeded them vs. the scale-down model. The slight variation in titer between the two bioreactors resulted in slightly higher productivity in the second run than the first. Higher titer feedstocks can be purified with greater efficiency in an MCC process due to the reduction in load time to column saturation. The MCC process enables ~1 kg of product to be processed in under 3 hours.

The ability to use up to eight columns on the Octave PRO allows processing feedstocks with an even higher titer, reducing future downstream bottlenecks as titers continue to increase.

During the scale-up process, an important observation was made: the 1 kg MCC process maintained the same column diameter (14 cm ID) as the scaled-down 100 g batch process. The utilization of MCC allowed for the addition of more columns and increased process productivity, enabling the processing of significantly larger product masses, in this instance, a tenfold increase, while still maintaining smaller column dimensions. This advantage of MCC becomes particularly significant when considering the non-linear cost increase associated with larger prepacked columns compared to smaller column sizes.

Summary

The Octave BIO and PRO systems simplify the transition from a legacy batch purification process to an MCC process. Scaling up the process from a 100 g pilot scale to a 1 kg clinical scale on the Octave PRO was executed with the same PROComposer software tool that automatically scales the MCC process steps according to the mAb titer and column dimensions entered.

Compared to the reference batch process, the implementation of an Octave MCC system for this specific mAb capture process results in improvements in process productivity, resin utilization, and buffer consumption. These enhancements led to reductions of over 80% in resin volume and over 40% in buffer usage, respectively.

Overall, upwards of 1 kg mAb was produced in about 3 hours, all with only 4.71 L of resin. Catalent now considers MCC as an option of their process intensification offering alongside their GPEx® Lightning cell line development technology.

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