

Hybrid Approach to Continuous Downstream Biomanufacturing

The best of both worlds – Single Use and Multi-Column Continuous

Adoption of single-use (SU) technology to entire production schemes is only recently realized after decades of having disposable products available. The use of buffer and small disposable bioreactor bags were implemented early while more sophisticated or large unit operations employing SU have taken longer and are now available to complete the “disposable facility dream”. Likewise, the implementation of continuous production has begun in a stepwise fashion either by addressing the largest, individual unit operation bottleneck or by adopting the simplest, lowest risk technology to achieve early ROI and productivity gains.

Multi-column continuous chromatography (MCC) at production scale has been an ambition of GMP facilities for some time. Implementation of MCC at large scale greatly increases productivity, particularly at the capture step where many production facilities are bottlenecked as a result of productivity gains in upstream fed-batch or perfusion processes. The most advanced and simple MCC systems are available in stainless steel – not single use. Some of the drivers to maintain MCC systems in durable materials include low operating costs and enhanced performance gained by stainless steel (SS) metering pumps.

How do you gain the best of both the single-use and advanced SS component and manufacturing technologies? YMC America’s Process Technologies Group has a proven solution.

Single use environment enabled

A proprietary manifold enables the durable (SS) MCC unit (the YMC Contichrom TWIN LPLC) to automatically and aseptically manage multiple buffers, CIP solutions, and WFI from single-use bags and tubing sets. This arrangement allows use of the system in a minimally classified area, saving significant cost associated with more highly classified cleanroom production environments. The adaption to a commonly available single use connector that allows for sterile connect, disconnect, and reconnect simplifies the interface between the single-use and stainless-steel components on both the upstream and downstream sides of the unit. The ingenious design and automation assure the sanitization of all product-facing surfaces prior to opening the SU connector assembly resulting in a fluid path that is never exposed to an open environment that is not sanitized.

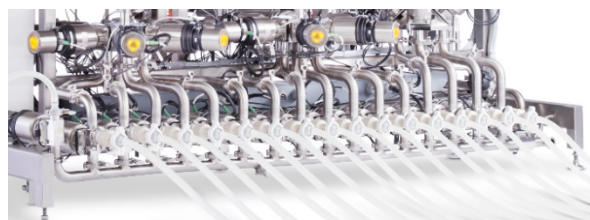


Figure 1: A proprietary buffer, feed and CIP manifold maintains an aseptic, closed environment between single use equipment and the YMC Twin system.

Employed on both batch and continuous chromatography systems, YMC has evolved the design first implemented on multiple production-scale systems in 2012 for operation in GMP environments (flow rate, ~30 L/min).

The most recent deployment of the manifold was on several multi-column systems provided to a GMP manufacturer of monoclonal antibody (mAb) based therapies. This design accommodated 16 buffers as well as WFI and NaOH from single-use bags. The user was able to operate the stainless steel-based multi-column chromatography system in a lower classification manufacturing suite (flow rate to 20 L/min).

Hybrid approach extends to unit processes

Incorporating MCC does not require that the entire process train be capable of continuous or extended batch operation. The implementation of the YMC Contichrom TWIN LPLC unit into a fed-batch scheme achieves the desired de-bottlenecking and does not require other unit operations to immediately be qualified for continuous production. This again is a demonstration of a hybrid unit process where both SU assemblies and SS systems are used. This stepwise approach to implementing continuous processing one unit operation at a time increases confidence and reduces risk while delivering productivity increases and substantial cost benefits.

Flexibility and intensification with novel platform features

MCC by itself substantially intensifies the capture process with productivity gains of nearly 3X (1). The YMC manufactured system not only provides a simple 2-column method for continuous capture (a patented process by YMC ChromaCon® AG), it also combines 4 additional unit operations in a single skid. The multi-function capability enables process intensification that significantly enhances productivity, while minimizing facility footprint, increasing asset utilization, and reducing cost.



Figure 1: Successful scale up of 100x on 2-column continuous capture systems from bench to GMP pilot.

Shown: Contichrom CUBE benchtop and YMC Contichrom TWIN LPLC GMP pilot.

A single YMC Contichrom® TWIN LPLC system can perform:

- Single-column Batch – with an outstanding range from 0.4 to 20 L/min
- Continuous capture – patented CaptureSMB® 2- column technology
- Sequential (Integrated) polishing – continuous 2-step chromatography
- Buffer in-line dilution (BID) – on-board in-line dilution of buffers and CIP solutions
- Single use aseptic interface – permits use in minimally classified suite

Multi-functional systems are being deployed in GMP manufacturing to deliver flexibility in operations and to intensify several process steps to increase productivity.

Simple, robust system design

Continuous, multi-column Protein A capture chromatography is shown to debottleneck the downstream purification of monoclonal antibodies. The advantages of this type of continuous process as compared to traditional single-column capture include improved resin capacity utilization, shorter processing times or reduction in resin volume, and reduction in buffer consumption, all while maintaining yields at >98%. However, achieving these benefits comes at the expense of increased hardware complexity and investment. More complex equipment also increases risk of downtime and maintenance costs.

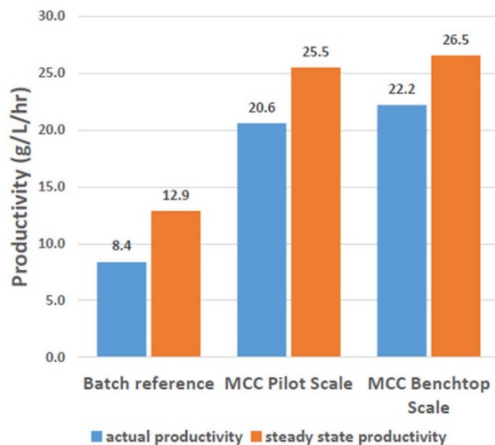


Figure 2: A 2- to 3-fold increase in productivity when using continuous capture over single-column batch (2).

While systems and processes utilizing up to as many as 8 or more columns have been reported, a simple 2-column continuous system maximizes process performance with respect to resin capacity utilization, buffer consumption, and product concentration while minimizing hardware investment and risk (3). Fewer columns significantly reduce equipment complexity simply by reducing the number of valves, plumbing, and monitoring devices required. Simpler hardware also means simpler operation and automation facilitating operator

understanding, equipment qualification, and process validation.

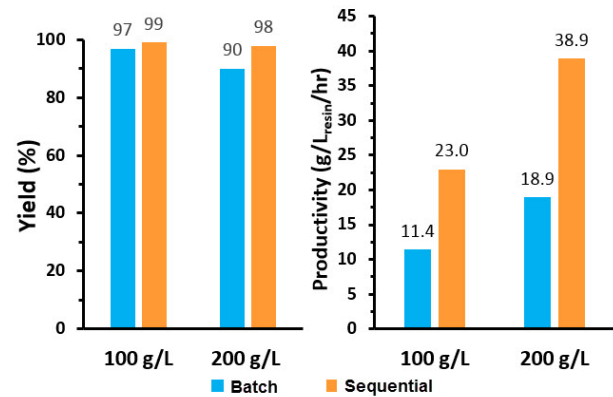


Figure 3: Data for two different loads shows a consistent twofold increase in productivity when using sequential over batch (2) while maintaining a high yield.

Sequential batch operation

The continuous operation of two interconnected downstream unit operations (e.g. flow through and bind-elute polishing steps) is another way to increase productivity and intensify the downstream process. Two-fold increases in productivity for continuous, sequential batch operation as compared to individual batch steps has been reported (2). In addition, operating two, connected polishing steps continuously eliminates the need for in-process hold and storage and eliminates the time associated with changeover between individual unit operations. Operating sequential batch operations continuously on a two-column system, such as the YMC Contichrom TWIN LPLC, combines two unit operations in one system, reducing the equipment footprint to maximize throughput per square foot of operating suite.

System optimized for use in a single use, buffer concentrate environment

The YMC Contichrom TWIN LPLC with in-line buffer dilution option (4) provides:

- Accurate and reproducible dilution delivers consistent point-of-use buffer every time
- Use of buffer concentrates – saves space
- Dilution of concentrated caustic – for fully automated system CIP / sanitization
- Facilitates sequential processing - creates in-line buffer for conditioning of feed between sequential two-column steps
- Buffer on-demand enhances scheduling flexibility
- Significant space savings – two unit operations combined and use of concentrates further decreases tankage by 10X or more

De-risk the continuous challenge and gain significant flexibility and productivity

Using this uniquely simple, patented design eases production implementation, validation and maintenance. The new YMC TWIN enhanced platform delivers scalable, proven cost reductions (>50% Protein A and buffer reduction) (4) significantly increased production flexibility, 2-3-fold productivity gains and the ability to combine multi-unit operations on a single skid in a single-use environment. Simple design with options for multi-function enhancements de-risks the investment both in validation ease and manufacturing asset utilization. This is truly the Swiss Army Knife for modern biomanufacturing facilities.

Wide range in commercial production

Units with the functionalities mentioned above are now commercialized with a flow ranges of up to 20 L/min, a platform enabling small to large production on the same system.



Figure 4: YMC Twin with batch, continuous capture (CaptureSMB), buffer in-line dilution (BID) and sequential polishing capabilities. Also shown is the optional single-use buffer management manifold.

(1) James Angelo, John Pagano, Thomas Müller-Späth, Kathleen Mihlbachler, Srinivas Chollangi, Xuankuo Xu, Sanchayita Ghose, and Zheng Jian Li, Scale-Up of Twin-Column Periodic Counter-Current Chromatography for MAb Purification, BioProcess International Vol. 16(4) April 2018

(2) James Angelo, Srinivas Chollangi, John Pagano, Daniel Baur, Kathleen Mihlbachler, Thomas Muller-Spath, Xuankuo Xu, Massimo Morbidelli, Sanchayita Ghose, Scale Up Design and Optimization for an Intensified Downstream Process Utilizing Multi-Column Operations, 255th ACS National Meeting, New Orleans LA, March 2018

(3) Daniel Baur, Monica Angarita, Thomas Muller-Spath, Fabian Steinebach and Massimo Morbidelli, Comparison of Batch and Continuous Multi-Column Protein A Capture Processes by Optimal Design, Biotechnol. J. 11(7)2016:920-931.

(4) Mary Jo Wojtusik, Kurt Wilner, Low Pressure Liquid Chromatography Using EcoPrime LPLC with Enhanced Buffer In-Line Dilution, BioProcess J.,15(3)2016:14-19



Figure 5: YMC TWIN is also in operation using pre-packed disposable chromatography columns.

Editor's note: This technology was originally introduced by LEWA-Nikkiso America Inc. In late 2018 YMC Co., Ltd acquired the technology, manufacturing site, staff and license from LEWA. YMC now produces the Contichrom TWIN as well as the scale-down Contichrom® CUBE bench instrument by YMC ChromaCon AG).

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