MAGic™ BioProcessing

Large-Scale Magnetic Separation for the Processing of Monoclonal Antibodies

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The work presented in this paper demonstrates that the novel large-scale magnetic separator, MAGic™Accio PROCESS, developed by MAGic Bioprocessing has the potential to transform antibody manufacturing as it is known today. The system integrates upstream (USP) and downstream processes (DSP) and, through its magnetic bead–based separation method, eliminates the need for clarification and filtration steps. To assess the economic feasibility of such a magnetic separation-based process, a study based on a comparative economic model was conducted in comparison to the conventional monoclonal antibody (mAb) production process. The study revealed that integrating cell removal and product capture in a single operation is the main factor driving the unified productivity between USP and the magnetic bead-based processes. This results in significant economic benefits, including reductions in cost of goods per gram of mAb and providing an overall increase in annual facility output.

Introduction

The use of monoclonal antibodies (mAbs) as therapeutic has increased dramatically over the past few decades. To meet the growing demand for mAbs and other antibody-based therapeutics, manufacturing processes are continuously being optimized to enable the production of larger quantities at reduced cost.[1,2] These increasing demands have driven significant advancements in upstream processing (USP), leading to remarkably high monoclonal antibody (mAb) titers in mammalian cell cultures, up to 10 g/L in fed-batch cultures[3] and even greater productivity in perfusion-based systems. However, these improvements place substantial pressure on downstream processing (DSP), which must efficiently handle the resulting high product loads, increased cell mass, and overall rise in volumetric mAb productivity.[4] This impacts subsequent the clarification, capture and purification steps, e.g., with more challenging cell removal. Hence, DSP needs to evolve in sync with the USP productivity.

In recent years, many manufacturers have shown growing interest in adopting disruptive technologies and conducting studies aimed at supporting decisions to modify or replace their legacy process platforms, an essential step to address the emerging challenges in biopharmaceutical production.

Chromatography is a cornerstone of modern downstream processing, widely used for the separation and enrichment of macromolecules. Traditional bioproduction processes, such as mAb manufacturing, rely heavily on column chromatography[5], where clarified culture broth is required, as solid particles, such as cells and cell debris, can cause major interferences during chromatographic operations. Therefore, clarification steps become indispensable in legacy processes. Centrifugation and depth filtration steps are commonly employed to remove solids prior to column-based product capture.[5] However, these operations add significant cost and complexity, while their sole function, removing solids, does not directly contribute to product concentration or purification. As a result, clarification remains one of the major bottlenecks in current bioprocessing workflows.

To overcome this limitation, magnetic separation, long used at laboratory scale, has been investigated as a promising alternative.[6] Magnetic separation can handle high cell density in the load stream, is gentle on cells leading to lower release of impurities and offers rapid processing.[6] Integrating magnetic separation into biomanufacturing offers several advantages. By consolidating multiple operations into a single step, it reduces process complexity, conserves resources, increases yield, and enhances overall efficiency in a sustainable manner. For monoclonal antibody production in particular, where demand

continues to rise alongside cost pressures, such a technology represents a compelling advancement. However, the absence of a truly scalable magnetic separator has been one of the main barriers for its implementaion at large scale. This gap has now been addressed with the introduction of the world's first large-scale magnetic separation, the MAGicAccio PROCESS. The technology is based on permanent magnetism, enabling true scale-up instead of scale-out limitations typically associated with electromagnet-based systems. In its largest configuration, the MAGicAccio PROCESS accommodates up to 40 L of magnetic beads, is fully automated, and integrates smoothly into existing production workflows.

This study investigates the cost-competitiveness and the feasibility of modelled magnetic separation-based DSP scenarios addressing the challenges imposed by cell culture intensification on DSP steps, at industrial scale. A mAb manufacturing campaign of a total of 60 batches in one year was modelled based on two MAGicAccio magnetic separation processes compared to the legacy process employing centrifugation and filtration for cell removal prior to column chromatography for the target capture. The study focuses on the economic understanding and implications of the two magnetic-based separation processes relative to the legacy process. Moreover, the objectives were to assess the impact of the integrated cell removal and target capture, provided by the magnetic separation, regarding the major cost drivers and their contribution to cost of goods per process step and to facility output. The comparison of a magnetic-based separation manufacturing process to the traditional column-based process offers an in-depth understanding of the trade-offs between the different technologies and identifies the key parameters that influence the selection of the best and most cost-effective manufacturing strategy.

Materials and Methods

Manufacturing scenarios - The process economics analysis presented in this study is based on three distinct DSP scenarios. Namely, the traditional process (CHROM), a magnetic beads-based process with two polishing steps as in the CHROM process (MAG-2) and a similar process with only one polishing step (MAG-1), see Figure 1. The upstream process design is identical for all scenarios (see model assumptions). In the CHROM process (Figure 1 A) harvest clarification is achieved using a disk-stack centrifuge, where biomass-dependent yield is assumed, followed by a filtration step to remove remaining solids, both resulting in yield losses. The clarified broth the proceeds through Protein A capture, low-pH virus inactivation, two polishing chromatography steps, virus filtration, and finally ultrafiltration/diafiltration (UF/DF). This standard mAb downstream configuration serves as the basis of the economic analysis. Two alternative DSP strategies replace the traditional clarification and target capture with magnetic separation using a large-scale magnetic separator, such as the MAGicAccio separator. In the MAG-2 process (Figure 1 B), centrifugation, filtration, and Protein A capture are replaced by a single magnetic separation step capable of processing non-clarified broth, thereby integrating clarification and capture. All subsequent steps are identical to the CHROM process. The MAG-1 process (Figure 1 C) further refines this strategy by removing one polishing step, supported by bench and pilot scale results indicating low mechanical stress and correspondingly low host cell proteins (HCP) levels, which are assumed to justify eliminating this step while maintaining product quality.[6, 7]

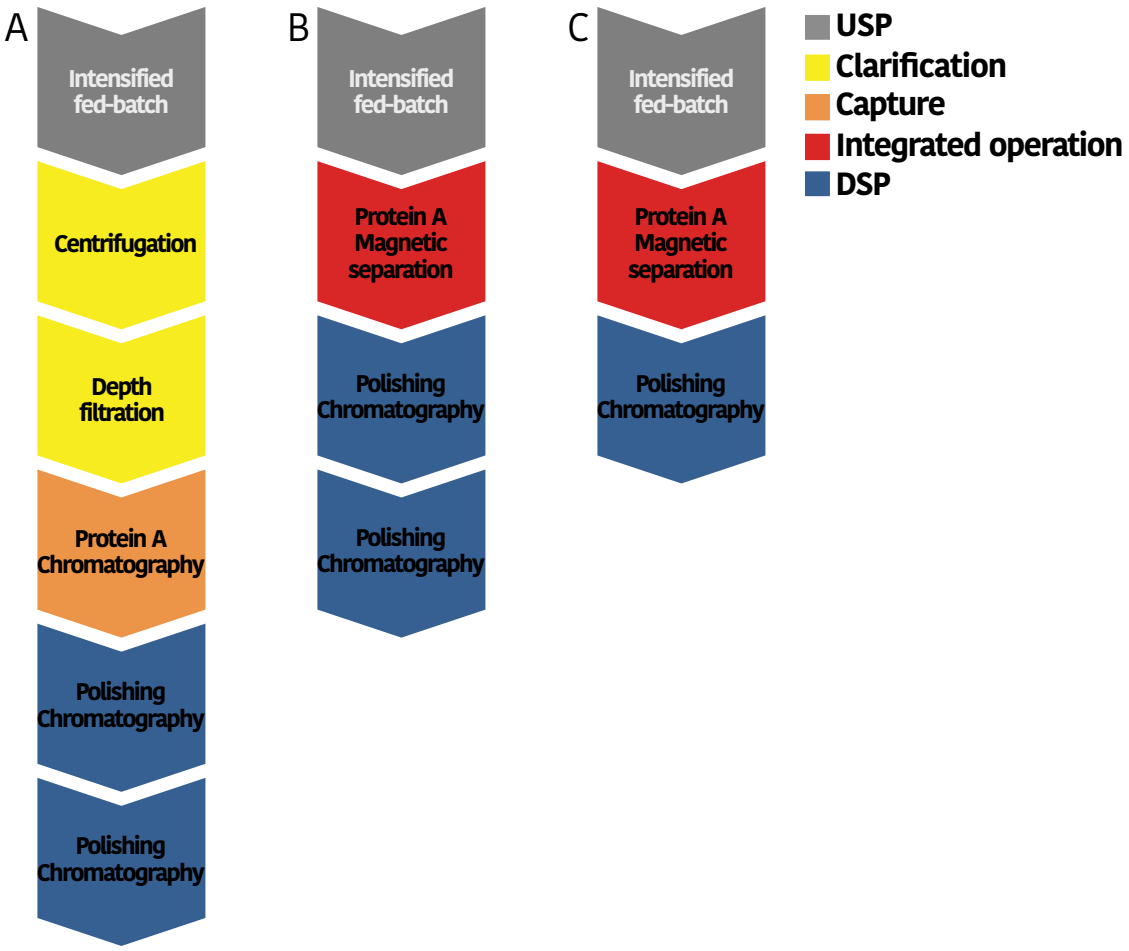

Figure 1. Flowsheets for the three modelled processes: (A) CHROM, (B) MAG-2, and (C) MAG-1 show the relevant operational steps. Post-capture DSP also includes UF/DF and two viral safety steps not depicted here: low-pH virus inactivation (VI) and virus filtration (VF).

Model assumptions - The USP is modelled with 4 bioreactors operated in staggered mode. For the cost of goods breakdown model, the bioreactor volume was determined at 500 L with two different titres: 6.6 g/L and 10.8 g/L. For the overall process output model, varying bioreactor scales (200 L, 500 L, 100 L and 2000 L) and mAb titres (3 g/L, 6 g/L, 9 g/L, 12 g/L and 15 g/L), representing variations in cell density were assumed. Each reactor is modelled to run 15 batches per year, with each batch lasting for 29 days in total This includes seed expansion for 15 days (3 times 5 days) and 14 days for the production bioreactor run. Hence, the model calculates a total of 60 batches per year (335 days of operational time) to maximize unit operation and facility utilization. Apart from the defined set points for the USP, the starting point for the tool is the mass balance calculation to assess the correct sizing of manufacturing equipment.

Calculations - With the given harvest volume and product concentration, the model calculates dependent parameters such as bead/resin volume (1), number of separator units (2), and magnetic separation phase durations (3, 4, and 5).

$$V_{beads} = \frac{Mass_{inlet}\,[g]}{DBC\,[\frac{g}{L}]} \tag{1}$$

$$n = \frac{V_{beads}\,[L]}{Capacity\ Separator\ Unit\,[L]} \tag{2}$$

$$t_{load} = \frac{V_{broth}[L] + V_{beads}[L]}{n * Q_{load}\,[\frac{L}{h}]} \tag{3}$$

$$t_{wash} = \left(\frac{BV_{wash} * V_{beads}[L]}{n * Q_{wash}\left[\frac{L}{h}\right]} \right) * wash\ cycles \quad (4)$$

$$t_{elution} = \left(\frac{BV_{elution} * V_{beads}[L]}{n * Q_{elution}\left[\frac{L}{h}\right]} + t_{incubation}[h] \right) * elution\ cycles \quad (5)$$

Results and Discussion

Cost of Gods Breakdown - Figure 2 presents the step-related cost contributions of the different unit operations per produced amount of mAb for the three downstream scenarios: CHROM, MAG-2, and MAG-1. These unit operations are categorized into USP (grey), Clarification (yellow), Capture (orange for traditional chromatography and red for magnetic separation), and Post-Capture DSP (blue). All three scenarios are based on the same USP input at two titres: 6.6 g/L and 10.8 g/L.

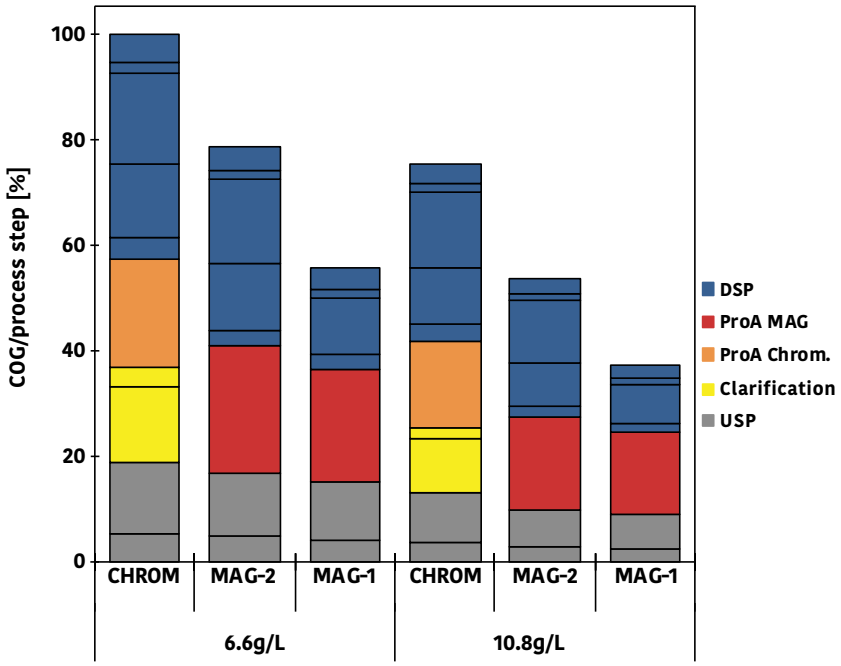

Figure 2. COGs contribution in percentage per process step across the three processes (CHROM, MAG-2, and MAG-1) at two different titers in 500 L scale.

When examining the total process-associated costs, a clear trend emerges: both magnetic processes (MAG-2 and MAG-1) show a reduction in COGs compared to the CHROM process. Depending on the titre process-related cost of gods are reduced by 29 to 38 % (Table 1). Integrating multiple unit operations into a single step, results in fewer steps and reduced step-related losses. This benefit is particularly evident when comparing the combined cost contributions of the Protein A-associated and clarification steps (Figure 2, yellow and orange). The CHROM process shows a significantly higher economic burden than the two magnetic-based processes (MAG-2 and MAG-1), with process-associated COGs for CHROM being nearly double. This highlights the economic benefit of step integration into a single operation. Such integration improves overall process yield and productivity, as more product is recovered from the same reactor input.

Table 1. Process performance for the three modelled processes at two different product titres in 500 L scale.

	6.6 g/L			10.8 g/L		
	CHROM	**MAG-2**	**MAG-1**	**CHROM**	**MAG-2**	**MAG-1**
Total COG/g change[1]	0 %	-18 %	-29 %	0 %	-27 %	-38 %
Output change[1]	0 %	15 %	20 %	0 %	34 %	40 %

[1]all changes are in relation to the CHROM process

Modelling of Annual Output - Complementing the cost of goods analysis, the annual output was modelled using different USP scenarios varying in scale, mAb titre, and cell density. The annual process output is shown as a function of USP titres of 3 g/L, 6 g/L, 9 g/L, 12 g/L, and 15 g/L, representing increasing cell densities, and across production scales of 200 L, 500 L, 1000 L, and 2000 L (Figure 3). The annual output model includes, as before, the traditional process and the two magnetic bead-based processes (MAG-2 and MAG-1), providing insight into how annual output scales with increasing USP titre.

For the traditional process (CHROM), the output curve shows a linear increase at low titres and cell densities; however, at higher titres the output deviates from linearity (Figure 3). The higher cell mass required to achieve these titres reduces the overall annual output of the CHROM process. This clearly highlights the bottleneck that is introduced by the cell clarification step, where increased cell mass, particularly during centrifugation, leads to poor yield. Although the titre increases fivefold to 15 g/L, the annual output rises only 3.85-fold, corresponding to a total process yield of 50% (Table 2). This demonstrates the strong negative effect of high cell densities on clarification efficiency, resulting in a significant product loss despite the increased USP productivity.

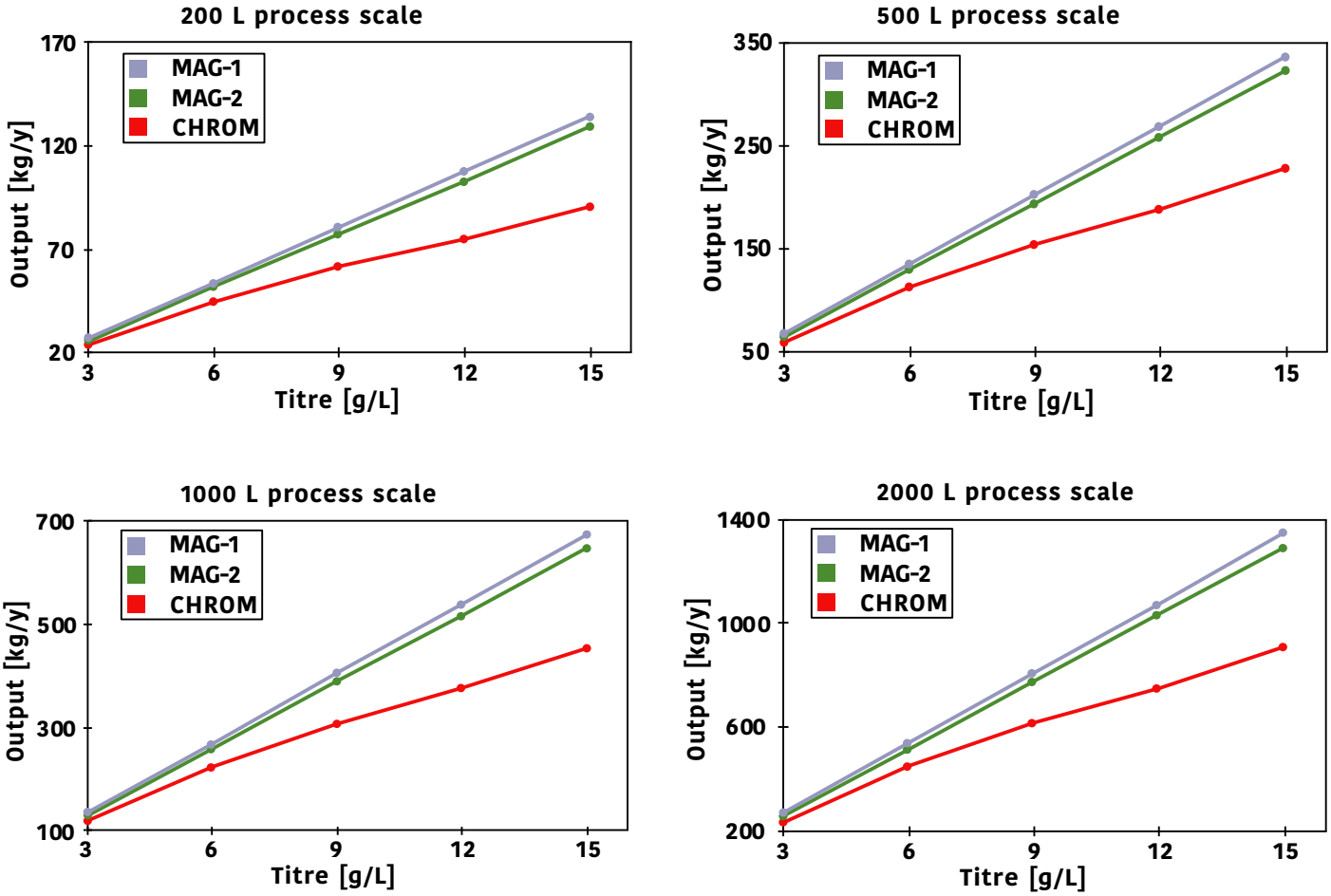

Figure 3. Modelled annual mAb throughput at titers ranging from 3–15 g/L for production scales of 200 L, 500 L, 1000 L, and 2000 L..

In contrast, the MAG-2 and MAG-1 processes are not negatively affected by cell density-related yield losses during clarification at any scale (Figure 3). Both magnetic bead-based processes show a linear relationship between increased USP productivity and annual output. For example, at the same fivefold increase in titre from 3 to 15 g/L, the magnetic processes maintain their yield without losses associated with that higher cell density. The overall process yields therefore remains constant across titres and cell densities, unlike the decline observed for CHROM (Figure 3 and Table 2). Additionally, for the MAG-1 process, the model assumes removal of one polishing step. Despite this reduction, the elimination of one polishing step does not lead to a significant gain in process yield (Table 2). This is because polishing steps typically exhibit high yields, and product loss during this phase is minimal. Hence, the integration of cell

clarification and capture remains the primary driver of process improvement, while polishing step removal contributes little to overall process economics.

In essence, the magnetic-based processes, through step integration and reduction, enhance overall process yield, leading to a higher product output, thereby improving the process related cost of goods contribution. When comparing the three modelled processes, both magnetic-based scenarios (MAG-2 and MAG-1) outperform the traditional CHROM process (see Table 1). They demonstrate significant gains in productivity and reductions in process related COGs. Moreover, the predicted process output (Figure 3 and Table 2) highlights the benefits of magnetic separation for processing high cell-density broths even further. Both MAG processes provide a stable yield across titres, whereas for CHROM, yield decreases with increasing titre. The model further demonstrates the clear limitations of traditional downstream processing (CHROM) in matching upstream productivity, with the cell clarification step representing a major bottleneck. As mAb titres and cell densities increase, clarification performance declines, leading to product loss, incomplete cell removal, and extended processing times. These losses have the most pronounced negative impact on overall productivity. Addressing this loss of productivity at high mAb concentrations and cell densities is essential to meet future production demands. These intensified conditions highlight the full potential of magnetic separation. Where centrifugation and filtration reach their limits, magnetic separation eliminates step-related losses and applies minimal stress to cells, thereby reducing the release of impurities such as HCPs. By comparison, conventional clarification methods impose high mechanical stress on cells, which increases HCP release[8] and consequently raises the purification burden.

Table 2. Modelled annual output at scale of the 500 L cell culture process in relation to harvest titre, USP output, overall process output and overall process yield.

500 L process scale	Harvest concentration [g/L]	USP Output [kg/y]	Process Output [kg/y]	Process yield [%]
CHROM	3	90	59	65
	15	450	227	50
MAG-2	3	90	65	72
	15	450	323	72
MAG-1	3	90	67	75
	15	450	337	75

Conclusion

Process intensifications of the mAb manufacturing have pushed the productivity of the USP side to exceptional high cell densities and mAb titres. However, current clarification technologies struggle to handle these high cell densities, resulting in poor yield and subsequent loss of productivity in DSP. Consequently, much of the gained improvements in USP is lost in the following steps.

This study demonstrates how antibody production processes using magnetic separation perform in a manufacturing context. The economic model provides valuable insight into how magnetic separation addresses the challenges of processing high-cell-density broths for mAb production. In particular, the integration of cell clarification and mAb capture into a single step is a key driver of performance improvement compared with the traditional process. Most notably, magnetic separation enables DSP productivity to keep pace with intensified USP, ensuring that upstream gains translate through the entire process.

MAGic BioProcessing has therefore developed the world's first truly scalable magnetic separation system, the MAGicAccio PROCESS, designed specifically for large-scale manufacturing of biologics based on magnetic separation, thereby enabling implementation of the process improvements and cost advantages predicted by the economic model.

Streamline the biologics manufacturing process with next-generation magnetic separation

References

1. Farid, S.S. Process economics of industrial monoclonal antibody manufacture. Journal of Chromatography B 848, 8-18 (2007).
2. Singh, N. et al. Clarification technologies for monoclonal antibody manufacturing processes: Current state and future perspectives. Biotechnology and Bioengineering 113, 698-716 (2016).
3. Handlogten, M.W. et al. Intracellular response to process optimization and impact on productivity and product aggregates for a high-titer CHO cell process. Biotechnology and Bioengineering 115, 126-138 (2018).
4. Mellahi, K. et al. Process intensification for the production of rituximab by an inducible CHO cell line. Bioprocess and Biosystems Engineering 42, 711-725 (2019).
5. Kelley, B. Industrialization of mAb production technology: The bioprocessing industry at a crossroads. mAbs 1, 443-452 (2009).
6. Brechmann, N.A. et al. Pilot-scale process for magnetic bead purification of antibodies directly from non-clarified CHO cell culture. Biotechnology Progress 35, e2775 (2019).
7. Brechmann, N.A. et al. Antibody capture process based on magnetic beads from very high cell density suspension. Biotechnology and Bioengineering 118, 3499-3510 (2021).
8. Pohlscheidt, M. et al. At Scale Analysis and Optimization of a Cell Culture Harvest Process– Mitigating Risk and Optimizing Performance. American Pharmaceutical Review (2013).

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