Enabling Process Simplification through Chromatographic Clarification with High Titer Cell Culture

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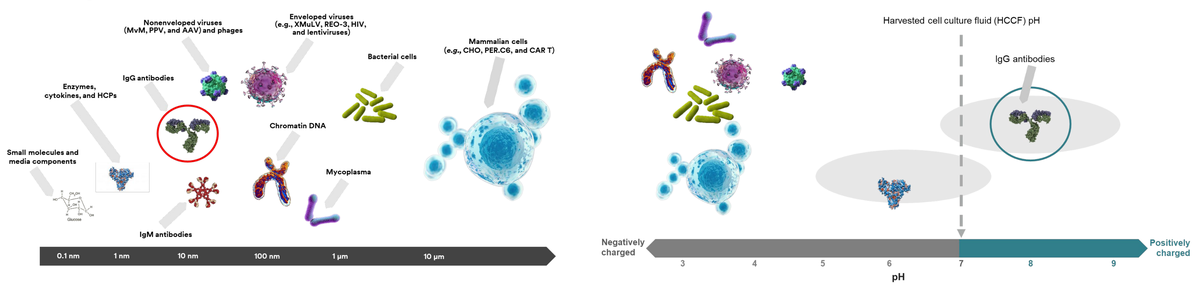
Introduction

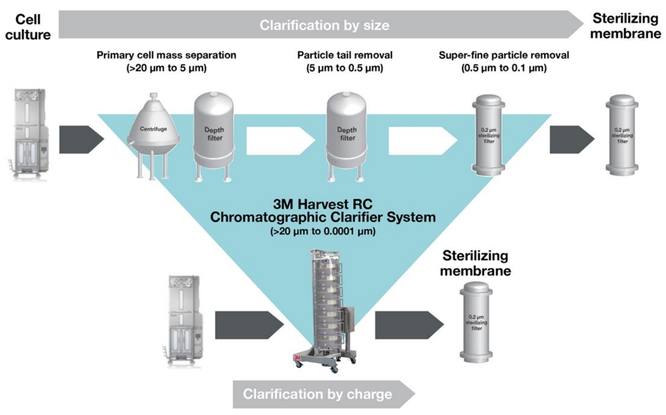
With advancements in upstream bioprocesses resulting in higher titer and cell densities, newer technologies has had to meet the demand for mass-transfer and mixing needs of an intensified process. However, as a new framework has been developed to support intensified cell cultures, newer challenges have emerged for the purification stages. The combination of higher cell levels along with process-related impurities (e.g., DNA/HCP/etc.) have posed a considerable challenge for downstream operations to meet the advancements that have been made in upstream bioprocesses.

Figure 1. Cell Culture Contaminants and Process-Related Impurities Plotted Along Their Size and Isoelectric Point.



3M™ Harvest RC is a harvest solution that utilizes fibrous anion exchange (AEX) chromatography to efficiently separate the cells, cell debris, and DNA from the harvest fluid containing the target product[1]. 3M™ Harvest RC is a new single-stage, single-use chromatographic clarification solution, designed as an efficient option to increase monoclonal antibody (mAb) yields while streamlining the upstream process by replacing the centrifuge and/or depth filtration process steps.

Figure 2. Process Compression using 3M™ Harvest RC Chromatographic Clarifier

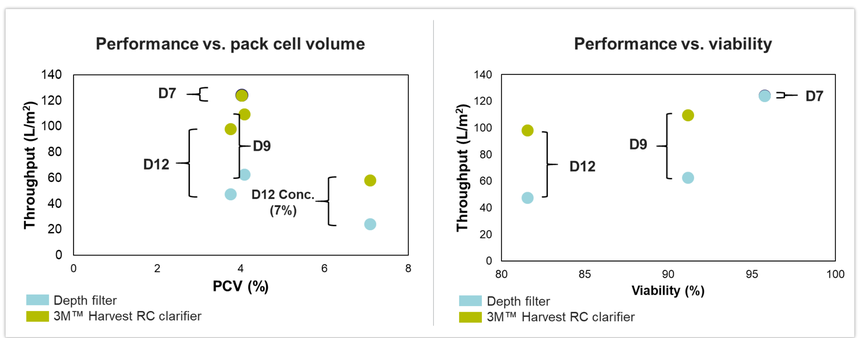


Results

Performance of 3M™ Harvest RC Chromatographic Clarifier vs. Traditional Depth Filtration

A cell culture was grown and harvested at different days to illustrate the impact of viability and pack cell volume on the 3M™ Harvest RC Chromatographic Clarifier and a single stage depth filter technology. The cell culture was harvested on day 7, 9 and 12 to illustrate the impact of viability on the performance of clarification. As viability dropped over the course of the culture run, material was harvested and challenged onto the two different clarification technology.

Figure 3. Throughput versus Pack Cell Volume for 3M™ Harvest RC Chromatographic Clarifier Performance compared to Traditional Depth Filtration Technology.



Robust Scalability of 3M™ Harvest RC Chromatographic Clarifier

Figure 4. Throughput Performance of 3M™ Harvest RC Across Different Scales

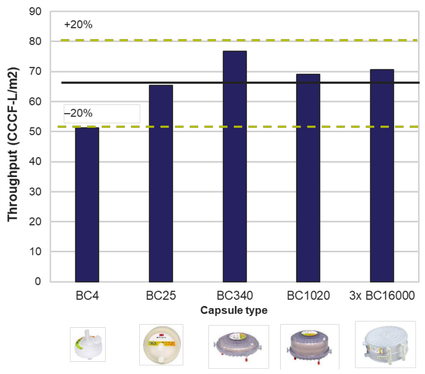
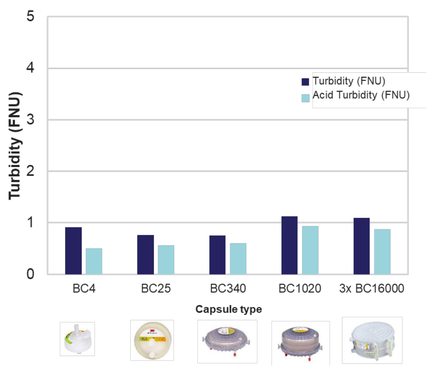


Figure 5. Turbidity Reduction using 3M™ Harvest RC Across Different Scales



Case Studies with Different Cell Cultures to Robust Performance

Different cell cultures were grown by Thermo Fisher Scientific to challenge the 3M™ Harvest RC Chromatographic Clarifier technology under a broad range of culture conditions. These cultures had diverse characteristics in cell density, viability, and pack cell volume. These different cultures were used to challenge the 3M™ Harvest RC as well as a traditional dual stage depth filter train from Supplier A. The results were then used to calculate the total surface area that would be needed to conduct a 2000L and 5000L scale run and the materials that would be needed.

Table 1. Four Different Cell Culture Characteristics for Challenging 3M™ Harvest RC Chromatographic Clarifier.

	Culture	1	2	3	4
Culture details at harvest	VCD (x 10^6 cells/mL)	27	20	15	9
	Viability (%)	91	86	83	76
	PCV (%)	12	14	8	4

Table 2. Total Surface Area Needed for Clarification at 2000 and 5000L Run Using A Traditional Dual Stage Depth Filtration Process versus 3M™ Harvest RC Chromatographic Clarifier.

Culture	1		2		3		4	
Train	Dual Stage Depth Filter	3M Harvest RC	Dual Stage Depth Filter	3M Harvest RC	Dual Stage Depth Filter	3M Harvest RC	Dual Stage Depth Filter	3M Harvest RC
2K filter area (m^2)	50 (total)	**29**	57 (total)	**41**	27 (total)	**19**	30	**14**
5K filter area (m^2)	123 (total)	**74**	142 (total)	**102**	66 (total)	**49**	74	**35**

Table 3. Case Study for Culture #3 for a 5000L Scale Run.

	Supplier A			3M™ Harvest RC	Improvement with 3M™ Harvest RC
	Stage 1	Stage 2	Total	Single stage	Single-stage reduction
EFA (m^2)	44	22	66	49*	**26% reduction**
No. of capsules	40	20	60	45	**25% reduction**
Footprint	–	–	11.4	7.6	**33% reduction**
Pre-use flush (L)	4,400 (100 L/m^2)	2,200 (100 L/m^2)	**6,600 L**	**1,225 L** (25 L/m^2)	**81% reduction**
Yield (%)		86		100	**14% increase**

Conclusion

- The 3M™ Harvest RC Chromatographic Clarifier can act as a single stage single use technology for promoting process compression through an innovative chromatographic clarification approach for capturing cells, cell debris, and soluble impurities.
- The results illustrated that the 3M™ Harvest RC had superior capacity compared to a traditional single stage or dual stage depth filter technology which can lead to reduced set-up time and waste generation.
- The use of the 3M™ Harvest RC could result in highly clean filtrate with a single stage that would result in a simplified process development and process set-up. This single stage set-up also resulted in improved yield compared to traditional multiple stage set-up that would translate to product loss.

References

1. Almeida, A., Chau, D., Coolidge, T., El-Sabbahy, H., Hager, S., Jose, K., Nakamura, M. and Voloshin, A., 2021. Chromatographic capture of cells to achieve single stage clarification in recombinant protein purification. *Biotechnology Progress*.

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