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Monoclonal Antibody Concentration | Diafiltration

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Abstract

This application note is focusing on mammalian cell clarification on the example of CHO cell clarification for a monoclonal antibody (mAb) process with a Single-use Line Explorer 12-inch (filter area: 155 cm²), 0.45 µm, 1.0 mm fiber ID. It was shown that for the target protein (140 kD) could be recovered nearly 100%.

Introduction

In the following you will see application data from a clarification of CHO cells during monoclonal antibody production using 2× concentration and 2 diavolumes.

Determine optimum membrane loading and permeate flux rate to maintain a stable and TMP ≤5 psig while maximum transmission of target protein (140 kD). Maximum transmission would result in high product recovery in shortest time.

Materials

For this mammalian cell clarification on the example of CHO cell clarification for a monoclonal antibody (mAb) process a Single-use Line Explorer Hollow Fiber Module was used. With a length of 24-inch and a pore size of 0.45 μm and 1.0 mm fiber ID. Like all our Hollow Fiber modules the membrane consisted of modified Polyethersulfone (m-PES). The Explorer module has a diameter of 1.3 cm and a corresponding filter area of 0.032 m²

Details of used Hollow Fiber Module

Family	Single-use
Product Size	Explorer
MWCO Pore Size	0.45 μm
Fiber ID	1.0 mm
Length	24 inch
Filter Area	0.0321 m ²
Crossflow rate ~ 4,000 sec ⁻¹ * [L/h]	25.8
No. of Fibers	18
Recommended batch volume per module	250 - 1,500 mL
Diameter Module (cm)	1.3 cm
Feed Retentate connectors	½-inch TC
Permeate connector	¾-inch Hose Barb
Material	SU94510EXP24S6 (6-pack)

Methods

In cell clarification of mammalian cell processes like this CHO cell clarification especially for antibodies, pore sizes between 0.2μm and 0.45μm are recommended.

Beside the recommended shear rate should be ideally between 2000-4000 sec⁻¹.

Feed:

- Total Cell Density: 1E07 cells/mL
- Viability: 90%

Details of Trial

Membrane & Module	Single-use Line Explorer 12-inch, 0.45 μm, m-PES, 155 cm ² , 1.0 mm fiber
Initial Feed Volume	477 mL
Membrane Loading	477 mL / 155 cm ² ≈ 30 Liters/m ²
Process Objective	12× concentration + 2 diavolumes (DV)
Process Flux (concentration)	Constant Permeate Flux at 30 LMH @ ~3,900 sec ⁻¹
Process Flux (diafiltration)	Constant Permeate Flux at 24 LMH @ ~3,900 sec ⁻¹

Results

The cell clarification of the monoclonal antibody from CHO feed was performed with a Single-use Line Explorer 12-inch (155 cm²), 0.45 μm, 1.0 mm lumen hollow fiber module. As shown in Figure 1, the transmembrane pressure profile at a constant permeate flux mode of 30 LMH was steady and ≤ 5 psig up to 2× concentration. During diafiltration volume 2, the buffer was not optimized for the product because the filtrate pressure increased >5 psig (see Figure 1). At a membrane loading of 30 L/m², the overall antibody recovery was nearly 100% as shown in Figure 2.

Volume Throughput Performance

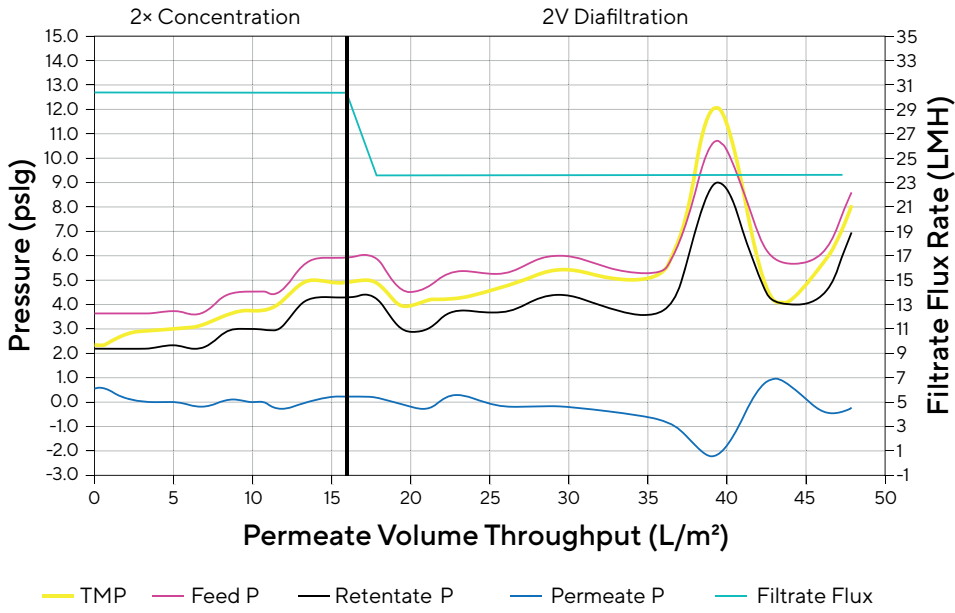


Figure 1: Pressure profiles during processing (concentration & diafiltration)

mAb Recovery (by Permeate Pool Mass Balance)

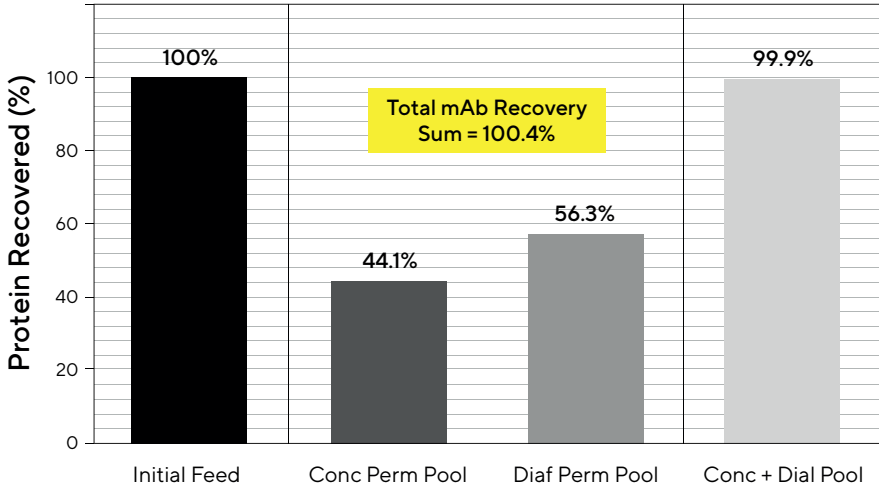


Figure 2: Monoclonal antibody (mAb) recovery by permeate pool analysis.

Conclusion

The optimum membrane loading was determined at 30 L/m² at a TMP ≤5 psig with an the overall antibody recovery of nearly 100% as shown in Figure 2.

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