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Application Note

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Recombinant Protein – Pilot Scale Concentration – 12.5 g/L

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Abstract

This application note is focusing on the concentration and diafiltration of a 150 kD recombinant protein. With a target concentration of 12.5 g/L.

The goal was to determine the optimum operating conditions to maximize the retention of the protein at a favorable permeate flux rate.

The Reuse line Investigator in 24-inch lengths and 30 kD MWCO with a 1.0 mm fiber ID was used for this trial.
I was shown that no passage of the recombinant protein into the permeate happened, therefore 100% of the product were retained. The process target of ≥10× concentration was even exceeded when the actual concentration was >15× with a processing time of 1.6 hours.

Introduction

In the following you will see application data from a concentration step of a recombinant protein production process. The goal was to determine an optimum membrane loading and permeate flux rate to maintain a stable and TMP ≤5 psig while maximum transmission of 150 kD target protein.

Materials

For this concentration and diafiltration of a 150 kD recombinant protein a Reuse line Investigator was used. The length of 24-inch and a MWCO of 30 kD and 1.0 mm fiber ID where chosen. Like all our Hollow Fiber modules the membrane consisted of modified Polyethersulfone (m-PES). The Investigator module has a diameter of 3.3 cm and a corresponding filter area of 0.28 m^2.

Details of used Hollow Fiber Module

Family	Reuse
Product Size	Investigator
MWCO | Pore Size	30 kD
Fiber ID	1.0 mm
Length	24 inch
Filter Area	0.28 m^2
Crossflow rate ~ 4,000 sec^{-1} [L/h]	240
No. of Fibers	160
Recommended batch volume per module	2 – 12 L
Diameter Module (cm)	3.30 cm
Feed | Retentate connectors	1.5-inch TC
Permeate connector	½-inch TC
Material	WA03010INV24S0 (1-pack)

Methods

Given the characteristics of the target protein (150kD) a Hollow Fiber Module with a MWCO of 30kD was chosen. The optimum operating conditions and maximum retention of the protein at a favorable permeate flux rate should be defined.

Feed:
- Recombinant Protein: 150 kD
- Concentration: 12.5 g/L

Details of Trial

Membrane & Module	Reuse line Explorer 24-inch 30 kD, m-PES, 0.27 m^2, 1 mm fiber ID
Process (target)	>10× concentration in approximately 2 hours
Process Condition	>14 LMH (4000 sec^{-1})

Results

During optimization, the flux vs. TMP (Figure 1) shows that process conditions were optimum for this recombinant protein. Figure 2 for filtrate flux vs. concentration factor shows the process was able to exceed to process targets of ≥10× concentration of the recombinant protein within 2 hours.

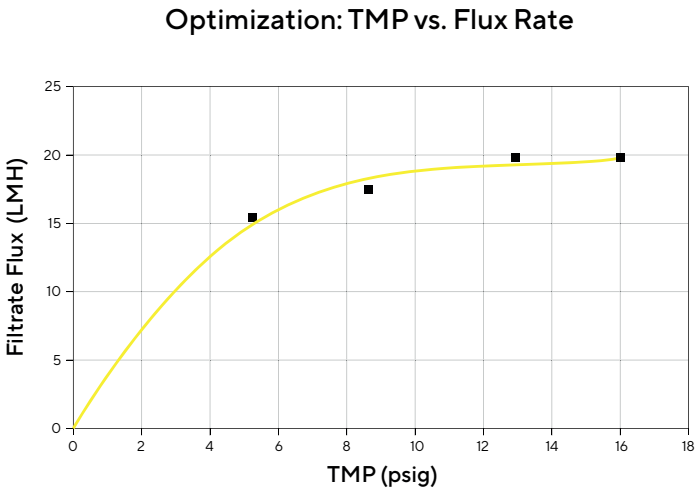

Figure 1: Filtrate Flux rate vs. TMP for recombinant protein

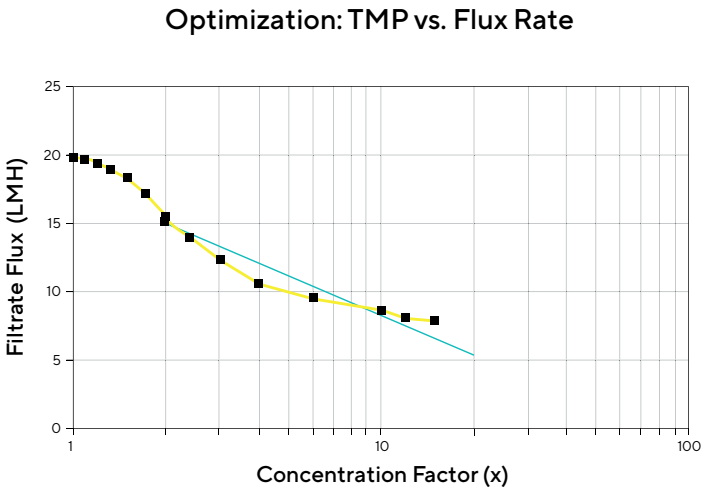

Figure 1: Filtrate flux profile for concentration of recombinant protein from 12.5 g/L to 190 g/L (>15× concentration)

Conclusion

The average permeate flux rate was 12 LMH during the concentration process. Using the 30 kD membrane showed no passage of the recombinant protein into the permeate, therefore retaining 100% of the product. The process target of ≥10x concentration was exceeded when the actual concentration was >15× with a processing time of 1.6 hours.

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