ABE 304
Bioprocess Engineering Laboratory

MEMBRANE SEPARATION FOR XANTHAN RECOVERY

INTRODUCTION AND SCOPE

Mechanical techniques such as filtration or sieving are used to separate components in mixture. The technique applied depends on the physical characteristics of the components in the mixture. A mixture of particles can go through a series of separation steps in an effort to concentrate or purify a single product. Separation processes are arranged to remove the largest particles first, then work down to purify the particle of interest, depending on the size. Techniques to remove large particles include sedimentation and centrifugation. Purification at the molecular level can include reverse osmosis or chromatography.

This lab will use a 500,000 NMWC ultrafiltration membrane to concentrate a 2.5% xanthan solution (Yang, Lo, & Min, 1996).

AIMS

- Determine the selectivity of the membrane in the process through the retention coefficient, R. There are two major types of membranes for ultrafiltration: cross flow or conventional. Figure 1 illustrates the filtration mechanism in place for each type of separation. The retention coefficient (Equation 1) is valid for both types.

$$R = \frac{C_R - C_P}{C_R} \quad (1)$$

where C_R is the concentration of the solute in the retentate and C_P is the concentration of the solute in the permeate at any point in the filtration process. Concentration is measured in g/L.

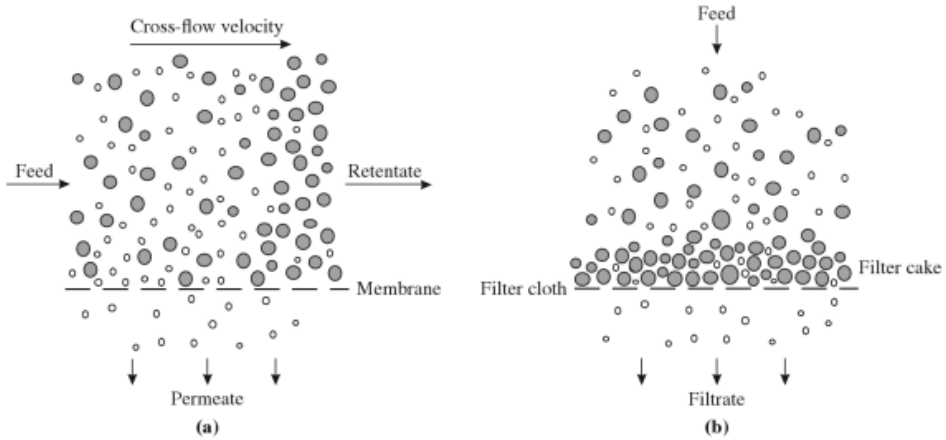

Figure 1: Types of membrane filtration (a) cross flow velocity and (b) conventional filtration. This lab will use the cross-flow type. Figure 11.23 (Doran, 2013).

- Determine the permeate flux, J (m^3/m^2s), as a function of the volumetric flow rate (Equation 2). Graphically relate the flux to the transmembrane pressure, ΔP (Pa) for each volumetric flow rate. Comment on the occurrence of concentration polarization if evident. Determine if the filtration occurs in the *pressure-controlled region*, or the *mass-transfer controlled* region.

$$J_v = \frac{F_P}{A} \quad (2)$$

where F_p is the volumetric flow rate of the permeate (m^3/s) and A is the area of the membrane (m^2).

- Determine empirical constants – a, b, & c – for the filtrate flux (Equation 4) and compare with literature values (Equation 5) (Lo, Yang, & Min, 1996).

$$J_v = 3.51 \times 10^{-5} \Delta P^a C_p^b Q^c \quad (4)$$

$$J_v = 3.51 \times 10^{-5} \Delta P^{0.25} C_p^{-0.64} Q^{0.32} \quad (5)$$

where Q is the pumping rate (m^3/s).

PROCEDURE

Peristaltic pump flow rate calibration:

The flow rate of the pump at different settings will need to be determined. It is conventional to list operations as the flow rate was set to 1.2 mL/min rather than saying the pump was set to 3. You will also need to know the pumping flow rate (m^3/s) for your analysis. The flow rate on the peristaltic pump is adjusted using the knob on the front of the pump drive.

1. Obtain a graduated cylinder, a container of water, and a stopwatch.
2. Use a free piece of flexible tubing to measure your flow rate. Do not flow through the entire system to measure the pumping flow rate.
3. Place the water at the pump inlet and the graduated cylinder at the outlet.
4. Turn the pump on by adjusting the knob.
5. Obtain the flow rate by either recording the time it takes to reach a specified volume, or by setting the time and recording the volume. Either way, the time and volume will need to be recorded.
6. Test flow rates between settings 3 and 7. The flow rates between the integer settings can be interpolated from a graph of pump setting (x-axis) versus flow rate (y-axis).
7. It is wise to test each setting at least three times to obtain reliable results.

Spectrophotometer calibration:

The spectrophotometer will need to be calibrated using a 0.1 g/L xanthan stock solution in the same fashion as was done during the fermentation lab. Refer to your lab notebook for the procedure. Zero the spectrophotometer with a 3:1 solution of H_2SO_4 to H_2O.

NOTE: One half of the group can calibrate the spectrophotometer while the other half calibrates the pump.

Filtration Operation

An ultrafiltration membrane will be used to concentrate the xanthan gum. Figure 2 shows the process for the experiment.

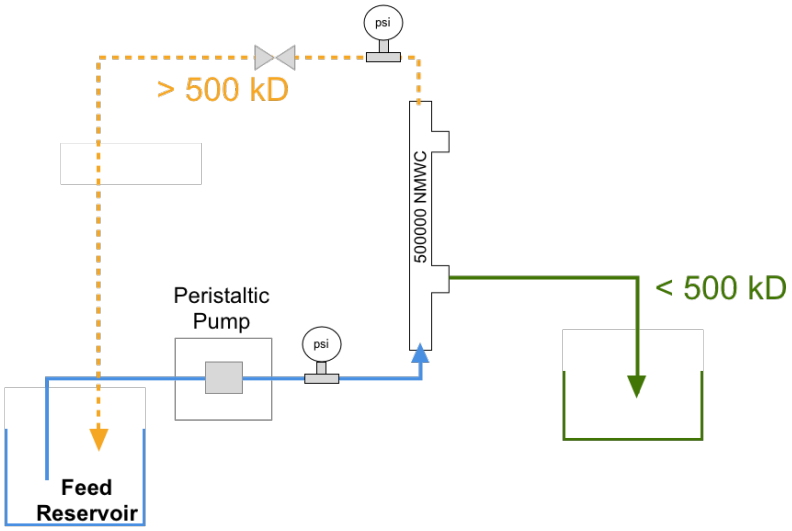

Figure 2: Schematic of membrane filtration process to concentrate phage from fermentation broth.

Membrane Operation

For this operation you will vary both the flow rate and the transmembrane pressure.

1. Remove the free tubing from the pump and place the membrane tubing back into the pump head.
2. Obtain your sample and place at pump inlet. The retentate reservoir outlet will be the same as the feed container. The reservoir should be placed on a heated stir plate set to 30˚C.
3. Turn the pump on to the lowest setting for your calibration.
4. Set the potentiometer to induce a transmembrane pressure of 2 psi. Wait until there is liquid flowing from each outlet into the specified reservoir before taking measurements.
5. Obtain a sample from each outlet and reserve for future analysis in the spectrophotometer.
6. Determine the flow rate of the permeate using a graduated cylinder. Use a stopwatch and a graduated cylinder similar to how you measured flowrate for the pump calibration. You only need to do this once.
7. You will need to repeat steps 4, 5, and 6 at transmembrane pressures of 4, 6, 8, and 10 psi.
8. **Before moving on to the next step**, reduce the transmembrane pressure to 1 psi and run the pump at the 5 setting for 3 minutes to flush the filter.
9. Change the pump flow and repeat the experiment until you have tested four flow rates at five transmembrane pressures. At each new setting, collect samples from the permeate and retentate, and measure the permeate flow rate. This should result in 2 sets of 20 samples.

NOTE: Take an initial concentration reading of your feed. Record the membrane surface area.

Xanathan Concentration

Test the concentration of xanthan in your solutions in the same manner as for the fermentation lab. Refer to your lab notebook for the procedure. You will need to perform a dilution to ensure that samples will be within range on the spectrophotometer. Choose a sample you think will have a high xanthan concentration and perform a 20x dilution. Test it on the spectrophotometer and adjust the dilution factor as necessary to make sure your reading is in the range of your calibration curve. When you have found a dilution factor that works, proceed to dilute and test the rest of your samples.

During each lab section, two groups will perform the experiments. Data from both groups will be pooled and should be used during data analysis. See shared spreadsheet in the Bioseparations folder in Blackboard.

DATA ANALYSIS

- Use the concentration of the solute in the retentate and in the permeate to determine the retention coefficient.
- Use a graduated cylinder and a stopwatch to calculate the volumetric flow rate of the permeate. Use the flow rate, along with the area, to determine the permeate flux.
- Use the transmembrane pressure for each flow rate and graph against the permeate flux. Determine if the membrane is operating in the pressure controlled or mass transfer controlled region.
- Determine empirical constants for filtrate flux from logarithmic plots of J_v, vs. ΔP, C_p, and Q. Compare with literature constants and comment if ΔP, C_p, and Q have a linear or non-linear effect on filtrate flux.

REFERENCES

Doran, P. M. (2013). *Bioprocess Engineering Principles*. Academic Press. Retrieved from https://books.google.com/books?id=wZSylDhgEXMC&pgis=1

Lo, Y. M., Yang, S. T., & Min, D. B. (1996). Kinetic and feasibility studies of ultrafiltration of viscous xanthan gum fermentation broth. *Journal of Membrane Science*, *117*(1-2), 237–249. http://doi.org/10.1016/0376-7388(96)00067-1

Yang, S. T., Lo, Y. M., & Min, D. B. (1996). Xanthan gum fermentation by Xanthomonas campestris immobilized in a novel centrifugal fibrous-bed bioreactor. *Biotechnology Progress*, *12*(5), 630–637. http://doi.org/10.1021/bp9600501

BONUS *(add to your appendix, refer to in the body of your report)*
Create a wire diagram for the lab. The wire diagram should include:

- Arduino
- 2x Cole-Parmer model 68075 pressure gauges
- Solenoid valve
- TIP120 transistor
- Sainsmart 1602 LCD keypad
- Potentiometer

The pressure gauge readings were inputs into the Arduino and the difference was compared to the pressure set by the potentiometer. If the conditional statement was true, the Arduino would send a PWM signal to the transistor to control the opening of the solenoid.