Modeling a batch foam fractionation process

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A mathematical model of a batch foam fractionation process has been developed based on analogies to a distillation model. The model is tested for the foam fractionation of the aqueous protein solution obtained from kudzu (Pueraria lobata) vine retting broth. This broth is most likely comprised of cellulases and pectinases. Time-varying trajectories of measured total protein levels are fitted to the model such that localized distribution coefficients (K’s) are determined at each stage, at 15-minute intervals, for the six-stage equilibrium model. This is the identification part of the model building. The distribution coefficients relate to the postulated local equilibrium between the rising foam stream and the descending liquid drainage stream. This is apparently the first time that such values have been determined directly from a dynamic foaming process. The K values are averaged to around 1.4 since they are approximately the same at each determined time point and position within the dilute protein solution (presumably governed by Henry’s Law). The simulation part of the model building uses the time averaged K’s to generate foamate (collapsed foam) protein concentrations which are compared with the sampled experimental data.

Introduction

Foam fractionation is to proteins what distillation is to solvents and hydrocarbons. It is a means of concentrating and separating hydrophobic proteins in a dilute solution from a bulk liquid phase to a foam phase. The main principles of the foam fractionation follow: One, the surfactant substances preferentially adsorb onto the gas-liquid interface (bubble surface generated by introducing a gas into the dilute protein solution through a porous media). Two, the bubbles rise through the solution and accumulate above the bulk liquid pool surface to form a foam phase with a small amount of liquid entrained in the thin films and Plateau borders formed between the bubbles. Third, this liquid tends to drain down the column and return to the bulk solution because of gravity, thus drying the foam phase. When the foam phase collapses, a concentrated solution (the foamate) remains. The drier the foam phase, the higher the resulting protein concentration. Proteins and en-
zymes are surface active due to the amphiphilic nature of these molecules. Foam fractionation is a low cost, initial purification method for dilute solution of biomolecules, which does not generally require the addition of expensive exogenous materials such as salts (which must be removed from the recovered product).

While numerous transport phenomena type of studies have appeared on foam fractionation of proteins (Uraizee & Narsimhan, 1990a, 1990b; Lockwood et al., 1997), little has been done to implement a stagewise countercurrent model to describe the column behavior. In the process modeled here, proteins being concentrated are from a liquid mixture obtained from the kudzu (Pueraaria lobata) plant retting process. In this process, kudzu vines are retted by anaerobic bacteria and fungi to loosen up the outer vine fibers and thus gain access to the desired strong cellulosic fibers, much like linen fibers are traditionally recovered from flax vines (Elamwat et al., 1998). The retting solution is comprised of soluble proteins, some of which can foam, such as cellulases and pectinases (Elamwat et al., 1998) which are potential high value by-products of the retting process. If the bulk liquid phase draining from the foam is denoted as the first phase, then the rising foam itself is the second phase and is analogous to the vapor phase in a distillation column. In the case of distillation, the vapor phase is generated from the original liquid phase by adding heat. In the case of foam fractionation the foam phase is generated from the original bulk liquid phase by adding a gas such as air. The liquid drained from the foam phase returns to the original bulk solution (undergoing bubble fractionation), and this recycling corresponds to the reflowing liquid in a distillation process (Lemlich, 1972). No exogenous surfactants or other non-gaseous additives are generally needed to this process to create the foam.

A mathematical model for this process is developed here to describe the time-varying total protein concentration profiles, as well as to determine distribution coefficients ($K$’s) between the foam and liquid drainage phases of the system. Since the $K$’s can vary with time, concentration and position and serve to simplify the model, as in distillation, they may only approximate equilibrium coefficients connecting the compositions in the adjacent two phases. Like the distillation process where data can be determined in batch, small-scale laboratory equipment under equilibrium conditions, foam fractionation is an inherently dynamic process that reaches equilibrium locally. It is assumed here that this local equilibrium is achieved rather quickly and in less time than the bubble resides in a defined separation stage. It appears like that because of the inherent nature of the process, foam fractionation “equilibrium” constants ($K$’s) are generally obtainable from the overall dynamic data using a dynamic model, although a small single stage process could be developed to simulate the equilibrium conditions. This paper focuses on both the identification step in which these $K$’s are determined by matching the model to the experimental concentration and flow rate data and the check step in which the foam concentrations are simulated from the average $K$ values and compared with different experimental data. If it should turn out that all of the $K$ values averaged to around the same value (in spite of the many approximations assumed here), then the $K$ value could be obtained from a single stage (very short) foam column for use in a multi-stage foam fractionation unit.

Material balances and equilibrium relationships are used to develop the model, much like in the development of distillation or absorption process models. The model describes the total protein concentrations of both the foam and the liquid drainage phases. The assumptions used to develop this model are chosen to be consistent with observations of the behavior of the column and measured variables and are invoked because there is no simplifying assumption, such as constant molar overflow used in distillation, to describe the foam drainage. At best there are empirical expressions. The measured variables are primarily the four effluent side stream protein concentrations and flow rates of the collapsed foam (foamate) phase, and two effluent side stream protein concentrations and flow rates of the bulk liquid. These samples are taken at six equally spaced positions along the axis of the foam fractionation column. It is assumed here that the height equivalent to a theoretical stage (HETP) is 0.2m, which is the spacing between sampling ports.

**Material and methods**

Kudzu vines of different diameters (0.3–0.7 cm) were harvested locally close to the intersection of Gay Street and 1st Avenue S. on the western bank of the Cumberland River in downtown Nashville, Tennessee, in July 1997. Approximately 1.3 kg of vines were cut at a time, washed, and placed in a 10-liter plastic container which was then filled with 7 liters of tap water (ca. pH 5.9) (Uludag et al., 1996) to give a solids concentration of 15.5 wt%. These cut vines of various lengths (13–16 cm) were kept submerged in that water at an ambient temperature of 23 ± 1°C. Water sampling was then performed every 1–2 weeks until retting was completed.
when the vines became loose and easily separated into fibers (ca. 2 weeks). A 400-mL sample filtered through Whatman No. 4 filter paper was used for each foaming experiment. The solution to be foam fractionated was adjusted initially to a desired pH by adding either 1 M NaOH or HCl prior to aeration.

Standard protein markers were from Sigma Chemical Co. (St. Louis, MO, USA). BSA: A-4378, MW = 66,000. Ovalbumin: A-7641, MW = 45,000. Cellulase: MVA-1284, was obtained from GIST-BROCADES, nV, Delft, Netherlands. Pectinase: P-9931.

The total protein concentration was assayed using the Coomassie Blue (Bradford) method ([Bradford, 1976] to measure the absorbance with a spectrophotometer. The linear relationship between the protein concentration and absorbance is:

\[ A_i = k_i C_i \]

(where \( A_i \), \( C_i \) and \( k_i \) are the absorbance, protein concentration and the conversion factor, respectively.) The conversion factor, \( k_i \), was determined by measuring the sample's absorbance and its actual protein concentration (taking a 1.0 mL sample solution and evaporating it in a hot air oven (ca. 45°C) for 10 hours and weighing it, and assuming the dry residue contained only proteins). The calculated conversion factor (\( k_i \)) of absorbance/concentration was about 1.81 l/g, when \( C_i \) was in g/L.

The foam fractionation experiments were carried out in a 1-meter long glass column (Fig. 1) with an inside diameter of 3.5 cm. A porous fritted glass sparger was fitted into the bottom of the column. The column included six sampling ports located along the length of its axis. Each port was covered with a rubber septum to allow for sample collection with a hypodermic needle. The ports were separated 20 cm from each other starting with the first port, which was located 2 cm above the sparger, as illustrated in Fig. 1. To minimize evaporation loss of water from the column, air was bubbled through a humidifier (Fig. 1) before it entered the bottom of the column. The airflow rate was monitored with a rotameter at 0.056 L/min, a typical aerobic fermentation airflow rate. Incidentally, successful use of this flow rate could lead to the on-line use of foam fractionation in conjunction with a foaming fermentation process. Only this airflow rate was used throughout this study. 1-mL samples (collapsed foam from the top 4 ports and liquid from the bottom 2 ports) were withdrawn every 15 minutes from each of the six side ports using a 10-mL hypodermic syringe throughout the experiment. Samples were taken by inserting the needle into the respective septum until it entered the bulk solution or the foam phase. The foam samples were collapsed using the syringe into a liquid foamate solution. The liquid samples were then dispensed from the syringe into 10 mm × 65 mm test tubes for subsequent protein concentration determinations. The protein assays included a total protein concentration for each sample, an occasional molecular estimation by gel electrophoresis and protein absorption spectral analysis. This spectrum was compared with spectra for one known cellulase and one known pectinase enzyme. The gel electrophoresis and spectral analysis indicated that two of the primary proteins in the kudzu solution mixture were consistent with the standard samples of cellulase and pectinase. They were only meant to qualitatively show that cellulase and pectinase could be present.

The total protein concentration was quantified spectrophotometrically at 595 nm using the Coomassie Blue (Bradford) method ([Bradford, 1976]) with a Spectronic 20 spectrophotometer (Bausch & Lomb, NY). The absorbance was measured after the Coomassie Blue dye was added to the sample, with a “zero” reference of deionized water under the same conditions.

Mathematical modeling

In distillation, heat provides the energy to create a vapor phase from a liquid phase ([Wanka t, 1998]), while in bubble/foam fractionation the gas (here, air) stream is used to create the bubbles for the bulk phase and the foam for the foam phase. The heart of the foam fractionation model, by analogy to a distillation model, is the postulated equilibrium between the two phases (one is the upward foam phase including bubbles and the liquid being dragged upward and the other is the downward liquid drainage phase) at each theoretical contacting stage. Here, that theoretical stage is a given distance along the column in either the bulk phase or the foam phase. That height is often called the HETP, the height equivalent to a theoretical stage (plate, in a plate distillation column) and is assigned to be 0.2 m here to be in keeping with the distance between the sampling ports. It is an approximation made to make this initial
study tractable. The distribution coefficient, \( K_n \), describing this equilibrium in the bulk or foam phases at a theoretical stage, \( n \), is defined by:

\[
c_n = K_n C_n \quad n = 1, 2, \ldots N
\]

(1)

where \( c_n \) is the protein concentration (in g/L) in the upward foamate (liquid) stream leaving the equilibrium stage \( n \), and \( C_n \) is the concentration (in g/L) in the liquid stream draining from equilibrium stage \( n \). In general, \( K_n \) varies with the position in the column as well as the operational time. It is a direct function of concentration and for dilute solutions it may approach a constant in keeping with Henry’s Law.

There are \( N \) stages (\( N = 6 \) for our model) in the bubble/foam fractionation column, as illustrated in Fig. 2. These stages correspond with the number of the sampling ports. The flow rate of foam (collapsed to foamate equivalent) leaving each port, \( f_{n,\text{out}} \), for \( n = 3, 4, 5 \) and 6, is established from experimental data. The effluent flow rate equals the collapsed amount of sample withdrawn from each port divided by 15 minutes, the time between sampling. This is a way to approximate a “continuous” effluent foamate flow term for the model by averaging a discrete measurement over the entire sampling interval time.

The effluent sampled values are then multiplied by a “flow factor” to give an estimated upward foam flow rate (on a liquid basis), \( f_n \), leaving a given stage \( n \). This approximation is in lieu of another modeling equation (analogous to the energy balance in distillation) to allow us to estimate the \( K \) parameter for this example. In sampling the foam phase, only the bubbles that are in an approximately spherical region surrounding the needle tip will be aspirated into the needle. Thus, a flow factor is needed to relate the sampled pencil-like foam rate to that of the entire upward foam flow (on liquid basis), \( f_n \), above a given stage. It was found that flow factor of ca. 550 was needed to convert the “measured” foam rate, \( f_{n,\text{out}} \), to the estimated foam flow rate, \( f_n \). This number is specific to the rate at which sampling occurs. This factor was established by simulating the model using various factor values until the slope of each of the simulated foam concentrations as a function of the column height, \( \frac{dc_n}{dn} \), became positive. In discrete form, this became \( c_N > c_{N-1} > c_{N-2} > c_{N-3} \), where \( N = 6 \). Only a narrow range of factors around 550 satisfied this criterion. If it can be established here that all of the \( K \)'s are nearly equal, then this approximation may not be necessary for future studies if the bottom \( K \) value can be observed from the initial foam “burst” measurements. Another way to avoid invoking this approximation is to use either a theoretical drainage equation from the literature or an empirical one developed for the particular example at hand (which will be done here).

The liquid (drainage) flow rates, \( F \)'s, were then determined by a stagewise balance,

\[
\frac{dV_n}{dt} = f_{n-1} - f_n + F_{n+1} - F_n - f_{n,\text{out}} \quad n = 3, 4, 5, 6
\]

(2)

where \( V_n \) is the volume hold-up of the net liquid (collapsed foam) on stage \( n \). The boundary condition at the top stage, (here, the sixth stage) is that the flow rate of the foam leaving the stage is set equal to “zero”. This physical constraint for the flow rate just above the \( N \)th (6th) stage follows because there is a physical barrier to flow at the top of the column, i.e., the column is “capped”. This is the condition of total reflux at the top. If the rate of change of net liquid holdup on stage \( n = 3, 4, 5, 6 \), or the left side of equation (2) is negligible, then the \( F \)'s can be solved from the above “steady-state” approximation of equation (2) with \( f_6 = F_7 = 0.0 \). The first two stages (\( n = 1 \) and 2) are combined into one single stage since they are both in the bulk solution phase and the
respective protein concentrations are nearly uniform, assuming complete mixing. There is a small amount of bubble fractionation in this section of the column but the amount of change is considered negligible here compared to the concentration changes in the foam part of the column. Here, the bulk solution phase is analogous to a reboiler in distillation where there is only one concentration at a given time due to the assumption of perfect mixing in that compartment. The stagewise mass balance for the bulk solution is:

\[ \frac{dV_b}{dt} = F_3 - f_2 - f_{out,b} \]  

Here, the term \( \frac{dV_b}{dt} \) cannot be neglected since mass is being removed from the column through the sampling ports.

The dynamic total protein balance is similarly written for stage \( n \) as:

\[ \frac{d(c_nV_n)}{dt} = f_{n-1}c_{n-1} - f_n c_n + F_{n+1}C_{n+1} - F_n C_n - f_{out,n}c_n \quad n = 3, 4, 5, 6 \]  

\[ \frac{d(c_bV_b)}{dt} = F_3 C_3 - f_2 c_b - f_{out,b}c_b \]  

The equilibrium relationship, equation (1), is substituted into equations (4) and (5) so that the \( K_n \)'s can be obtained when the other variables are known from experiments or calculated from equations (2) and (3). The steady-state assumption (the left side of equation (4) is close to zero), is also used to solve for the \( K_n \)'s using equation (4).

The above calculation procedure for the \( K_n \)'s is the identification step for the equilibrium constants using the complete multistage model. After the \( K_n \)'s are obtained, they are time averaged. Using these time averaged \( K_n \)'s, the protein concentration profiles in the foam are then simulated from equations (4) and (5) and the approximation \( f_n' = 550f_{out,n} \). It is noted that the sidestream sample concentration term most likely is a mixture of \( c_n \) and \( C_n \), thus creating another source of error in obtaining this estimate of \( K_n \). Note that the \( f_n \)'s are not constant, reflecting the fact that there is drainage and \( f_n < f_{n-1} \) for \( n = 4, 5, \) and 6.

**Experimental results**

**SDS-Polyacrylamide gel electrophoresis**

Fig. 3 shows the gel electrophoresis results of the markers and kudzu vine retting solution. The gel bands were drawn here since the original gel was too faint to reproduce for this paper. It is shown that known samples of cellulase and pectinase are consistent with the bands of the kudzu vine retting solution, implying that the retting solution is rich in cellulase and pectinase, as expected.

**UV-VIS spectra result**

Fig. 4 shows the UV-VIS spectra of standard cellulase and pectinase samples as well as the kudzu vine retting solution. This is another test for consistency as that demonstrated by gel electrophoresis using the standard proteins including BSA and ovalbumin, along with cellulase and pectinase as described in the caption to Fig. 3. That is, the kudzu vine solution apparently contains abundant cellulase and pectinase as expected for a retting solution, but does not, however, directly show the presence of cellulase or pectinase.
Table 1. The measured protein concentration results (g/L) for the pH = 7.0, initial solution concentration 0.15 g/L.

<table>
<thead>
<tr>
<th>Time/min</th>
<th>c₀</th>
<th>c₁</th>
<th>c₂</th>
<th>c₃</th>
<th>c₄</th>
<th>c₅</th>
<th>c₆</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.15</td>
<td>0.15</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td></td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>0.15</td>
<td>0.19</td>
<td>0.23</td>
<td>0.27</td>
<td>0.32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>30</td>
<td>0.14</td>
<td>0.30</td>
<td>0.38</td>
<td>0.49</td>
<td>0.61</td>
<td></td>
<td></td>
</tr>
<tr>
<td>45</td>
<td>0.12</td>
<td>0.49</td>
<td>0.62</td>
<td>0.76</td>
<td>0.86</td>
<td></td>
<td></td>
</tr>
<tr>
<td>60</td>
<td>0.11</td>
<td>0.40</td>
<td>0.56</td>
<td>0.65</td>
<td>0.77</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

The results of foam fractionation of kudzu vine retting solution

It is seen in Table 1 that the experimental protein concentration in the bulk solution, \( c₀ \), is gradually reduced in time as expected. This is due to the unsteady-state nature of the batch operation, where the foam protein results at the expense of the bulk liquid protein. While protein sampling does not significantly reduce the large volume of bulk liquid (Table 2) and its concentration, the protein concentrations in the foam phase are much more sensitive to change because their liquid volumetric holdup terms are much smaller than the liquid hold-up in the bulk solution. In the foam phase, the measured effluent foamate concentration increases along the height of column as shown in Table 1. This reflects the fact that the higher the foam stage, the drier the foam phase. This is a result of drainage, so proteins are more concentrated at higher positions in the column than at lower positions. For the same position, the foam concentration first increases up to a maximum value with time, and then decreases. The decrease is due to the removal of proteins from the column.

Modeling results

Table 3 gives the \( K \) values estimated from the model using the experimental flow rate and concentration results. It is seen that the time averaged apparent equilibrium constants are around 1.2 to 1.6. This indicates that the local protein concentration in the collapsed foam phase is 1.2 to 1.6 fold that of the corresponding drainage liquid concentration. The equilibrium constant of the bulk solution, \( K_b = 1.56 \), came from the foam concentration leaving the interface, which is determined from the boundary condition at the interface, and \( c_b = K_b C_b \). This determination of \( K_b \) can be made in the first few seconds of startup when the foam phase rises to only a few millimeters and it is then assumed that this \( K_b \) does not change with time. It is noted that for egg albumin the measured \( K_b \) for the same superficial gas velocity is about 8, reflecting the difference between protein solutions (Du et al. 2000). A 1.4 fold concentration average increase at each of the 4 stages could account for an increase of concentration from 0.15 g/L to 0.86 g/L and is, therefore, consistent with the data. Smaller \( K \) values at the higher positions were also estimates. This appears to be reasonable since the liquid concentration becomes larger at the top of the column and the surface protein concentration becomes closer to the liquid protein concentration, assuming a hyperbolic Langmuir adsorption

Table 2. Measured bulk liquid volume changes with time.

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>( V_b ) (mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>397</td>
</tr>
<tr>
<td>15</td>
<td>367.4</td>
</tr>
<tr>
<td>30</td>
<td>364.9</td>
</tr>
<tr>
<td>45</td>
<td>362.4</td>
</tr>
</tbody>
</table>

Table 3. The calculated equilibrium constants (K’s) from the model.

<table>
<thead>
<tr>
<th>Time/min</th>
<th>( K_b )</th>
<th>( K_3 )</th>
<th>( K_4 )</th>
<th>( K_5 )</th>
<th>( K_6 )</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>1.56</td>
<td>1.20</td>
<td>1.88</td>
<td>1.65</td>
<td>1.39</td>
</tr>
<tr>
<td>30</td>
<td>1.56</td>
<td>1.74</td>
<td>1.48</td>
<td>1.41</td>
<td>1.28</td>
</tr>
<tr>
<td>45</td>
<td>1.56</td>
<td>2.85</td>
<td>1.28</td>
<td>1.25</td>
<td>1.15</td>
</tr>
<tr>
<td>60</td>
<td>1.56</td>
<td>2.24</td>
<td>1.37</td>
<td>1.16</td>
<td>1.19</td>
</tr>
<tr>
<td>( K_v ) average</td>
<td>1.56</td>
<td>1.68</td>
<td>1.37</td>
<td>1.23</td>
<td>1.25</td>
</tr>
</tbody>
</table>

Table 4. Flow rates of the effluent foam and liquid drainage calculated from the model.

<table>
<thead>
<tr>
<th>Position, ( n )</th>
<th>( f_n )</th>
<th>( F_n )</th>
</tr>
</thead>
<tbody>
<tr>
<td>2(bottom)</td>
<td>4.582</td>
<td>–</td>
</tr>
<tr>
<td>3</td>
<td>3.667</td>
<td>4.553</td>
</tr>
<tr>
<td>4</td>
<td>3.056</td>
<td>3.652</td>
</tr>
<tr>
<td>5</td>
<td>2.619</td>
<td>3.046</td>
</tr>
<tr>
<td>6</td>
<td>0</td>
<td>2.614</td>
</tr>
<tr>
<td>7</td>
<td>–</td>
<td>0</td>
</tr>
</tbody>
</table>

\( f_n \) – Flow rate of the foam stream leaving equilibrium stage \( n \), mL/min. \( F_n \) – Flow rate of the liquid stream draining from equilibrium stage \( n \), mL/min.
isotherm (in which the $K$’s decrease as the concentration increases).

Fig. 5 shows the simulation results using the obtained average $K$’s. It shows consistency with the experimental results. Under the total reflux condition, $f_n \cong tF_n$ and $f_0 \cong F_0$, assuming steady state and negligible liquid withdrawn from the column. This is consistent with the calculated results (less than 1% error). The drainage results can be expressed by either (6) $F_n = F_{n+1} + \Delta n$, for $n = 3, 4, 5$, where $\Delta 3 = 0.901$, $\Delta 4 = 0.606$, and $\Delta 5 = 0.432$ or (7) $f_n = f_{n+1} + \delta + 1$, for $n = 2, 3, 4$, where $\delta 3 = 0.915$, $\delta 4 = 0.611$ and $\delta 5 = 0.437$, since $\Delta 5 \cong \delta 5$, $\Delta 4 \cong \delta 4$, and $\Delta 3 \cong \delta 3$, we can develop an empirical drainage equation for this system under these particular operating equations as:

$$\Delta n \approx \delta n = -0.239n + 1.6103 \quad (R^2 = 0.97) \quad (8)$$

Equations (6)–(8) can serve as counterpart to constant molal overflow in distillation and, if accurate, as a substitute for the invoked flow factor approximation.

Conclusions

Foam fractionation is an effective method of concentrating proteins from kudzu (Pueraria lobata) vine retting broth. The staged-model developed here is similar to the distillation model. The obtained apparent equilibrium constants ($K$’s) by fitting experimental data to the model are about 1.2 to 1.6. The simulation results are consistent with the experimental results. This is the first trial to develop a simple model for the protein foam fractionation and further tests on other systems are necessary to test its efficiency.

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References


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