Rate of Protein Absorption and Desorption on Cellulosic Ion Exchangers

Both equilibrium and rate studies were made for the adsorption and desorption of Bovine Serum Albumin (B.S.A.), Fraction *V*, from dilute solutions using a new cellulose based ion exchange resin. Using the equilibrium data and a simple two-film theory to describe the rate data it was shown that during the initial perioid of *adsorption* the rate was controlled by film diffusion, after which it was controlled by intraparticle diffusion. The *desorption* was largely intraparticle diffusion controlled except for a very short surface desorption step. It is suggested that the model could be used to describe other protein adsorption/ion exchange processes.

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SCOPE

Due to the complex nature of protein molecules and the change in the charge and structure of the molecule with the ionic environment, no general model for the rate of adsorption or desorption of protein molecules on ion exchange resins exists. A detailed study was made of the equilibrium and rate of adsorption and deadsorption of the protein, bovine serum albumin, for a cellulose based anion exchange resin. A simple rate model based on the two phase resistance theory was applied to both the adsorption and desorption. While the present model is clearly an oversimplification of the actual rate mechanisms, it allows the use of simple parameters that may be easily measured or estimated and has the great advantage of being simple enough to be used in predicting column kinetics and behavior such as breakthrough and resolution.

CONCLUSIONS AND SIGNIFICANCE

Using the cellulose based anion exchange resin trade name "Protion" (DEAE) the adsorption of bovine serum albumin (B.S.A.) under optimum conditions (no salt present, pH 6.5) was found to be very strong such that large amounts of the protein were adsorbed (300 mg/gm) at very low protein concentrations in the solution (0.1 mg/gm). For the desorption (3% salt, pH 10), the situation was reversed and even at high solution concentrations of protein the amount of protein in equilibrium on the resin was very small. By alternating between these conditions one could clearly use this as a method of concentrating or separating B.S.A.

More important from a fundamental point of view, the simple two phase resistance model was able to explain the adsorption and desorption rate data using a solution phase diffusion coefficient of B.S.A. of the order of magnitude to that in free solution ($D = 6 \times 10^{-7}$ cm^2/s) and a resin phase diffusion coefficient about (1/100) of the free solution diffusion coefficient. Also as predicted by the two phase resistance model and the equilibrium curves two distinct rate periods were observed for adsorption, one a fast solution film diffusion and the other a long resin phase diffusion controlled period. Similar results were observed for the desorption which however showed three distinct rate periods. Studies are underway to test the possible generality of this simple model using other resins and proteins and test its application for column operation.

BACKGROUND

There are very few published studies on the adsorption and desorption of proteins on ion exchange resin and no general model which could be used to estimate the rate process. Indeed due to the complex nature of process it may be impossible to develop any rigorous rate model. It was therefore decided to use a very simple but general rate model based on the two phase resistance theory which allows for both bulk solution diffusion and intraparticle diffusion. This model has been used to describe many processes such as solute transfer in gas absorption and liquid-liquid extraction, Treybal (1980), and diffusion of small ions in packed ion exchange resin columns, Helfferich (1962). Also there have been studies similar to the present one such as those of Furusawa and Smith (1974) and Colton, Satterfield and Lai (1975) of adsorption of large organic and inorganic molecules on porous media. These studies used more exact diffusional models than proposed in the present study and either assumed only intraparticle diffusion or in cases where bulk solution diffusion was coupled with intraparticle diffusion were unable to estimate the coefficients accurately. However both these studies found intraparticle diffusion to be controlling in many cases as was found in this study. Also the studies of Colton, Satterfield and Lai (1975) which was mainly a study of the intraparticle diffusion of polystyrene in porous glass cubes, included experimental studies of the intraparticle diffusion of two proteins cytochrome C and human hemoglobin which can be compared to results of this study.

EXPERIMENTAL

In the early 1970's Tasman Vaccine Laboratories Ltd., New Zealand marketed a cellulose based ion exchange resin trade named "Protion," similar to the cross linked ion exchange resins "Sephadex" or "Bio-Gel." The resin was made from regenerated cellulose with short chain lengths making it possible to use much higher flow rates through a column packed with "Protion" (50 mL/min-cm^2) than possible with the conventional resin. It was a weak-base (anion) exchange resin with diethylaminoethyl (-O-CH_2-CH_2-N(CH_2H_5)$_2$) groups and therefore referred to as DEAE resin. The resin is now manufactured and marketed by Phoenix Chemicals Ltd. a subsidiary of Waitaki New Zealand Refrigerating Ltd. Its new trade name is "Indion" and it is supplied in range of different forms and grades.

The properties of the resin used in these studies are summarized in Table 1. These studies included the experimental determination of the equilibrium and kinetic adsorption and desorption of bovine serum albumin.

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TABLE 1. RESIN PROPERTIES (IN THE OH^- FORM)

Resin Size:	
Kinetic Studies	r_o = 0.035 cm
Equilibrium Studies	r_o = 0.021 cm
Resin Density:	
1.1 gm (wet)/cm^3 (swollen resin)	
Resin Voidage:	35%
Resin Water Content:	72%
Small Ion Capacity:	1 meg/mL
pK of Ionic Sites:	6.5 and 9

Equilibrium Studies

The equilibrim data was obtained by gently shaking 0.2 gm (dry basis) resin with 300 mL of protein solution in a round-bottomed flask. Initially the resin was put through the usual pre-conditioning and lastly before beginning the equilibration experiment brought to equilibration with a solution of the same pH and salt concentration that was to be used in the protein equilibration. This preliminary equilibration was accomplished by passing the proper solution through a small bed of the ion exchanger for from 24 to 48 hours. Recently distilled water was used to reduce CO_2 and carbonates. The bovine serum albumin (B.S.A.) used was Sigma, Fraction V (96–99% albumin). Five milliliter samples were removed periodically (not more than 25 mL total) to analyze for protein by the micro-biuet method, Elleman (1962), which was capable of detecting protein concentrations down to 10 ppm and is reasonably insensitive to the type of protein analyzed for.

As no buffer was used, the pH was also checked periodically and for the studies at pH 10 the change in pH was always less than 0.5 units. For the experiment reported at pH 6.5, there was an increase in pH from 5.3 to a final pH of 6.5. The samples were equilibrated over a period of from 24 to 48 hours. A blank containing a mid-range concentration of B.S.A. but no resin was run as a check. Both the equilibrium and kinetic studies were at 20°C.

Rate Studies

For the kinetic studies, either 0.5 or 1 gm (dry basis) was agitated in a small stirred vessel with 100 mL of protein solution at 2500 rpm. The resin was pre-equilibrated as in the equilibrium studies at the same pH as was to be used in the experiment (pH 10) and 1 mL samples were withdrawn periodically (less than 10 mL total) for analysis of protein by the microbiuret method. The pH was maintained constant by addition of 0.1N HCl using a pH-stat for control. This was done in order to maintain simple boundary conditions but on further reflection was deemed undesirable. However, in all cases less than 0.1 meq. of HCl was added (giving less than 0.001M[Cl^-]) and most all this addition took place over the first few minutes.

In the case of desorption the resin was equilibrated at a pH = 10 but with no salt present.

THEORY

The basis for the kinetic model was taken as the simple two phase resistance theory, which assumes the overall resistance to diffusion may be approximated by two effective films, one in the bulk liquid phase surrounding the resin and the other in the resin itself. While it is true that this theory is only a very rough approximation to the actual diffusional process which must involve much more complex diffusional models like those described for ion exchange by Helfferich (1962), it is an extremely convenient approximation which due to its relative simplicity is often used in describing rates of adsorption or ion exchange in columns. When one begins to consider the diffusion of chemical species as complex as proteins, with many ionic charges varying with the ionic environment, it becomes impossible to apply completely rigorous models and it is necessary to start with a simple model that can be readily adapted for column operation.

In this model the flux is approximated by

$$N = k(C - C_i) = \bar{k}(\bar{C}_i - \bar{C}) = K(C - C^*) \qquad (1)$$

The first two expressions for the driving force in Eq. 1 represent the individual expressions for bulk solution film diffusion and for intraparticle diffusion with the respective effective mass transfer

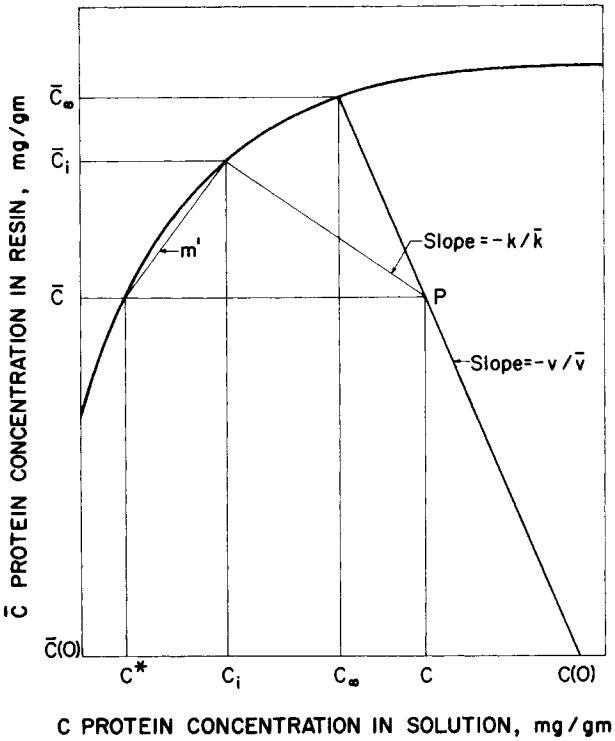

Figure 1. Typical absorption equilibrium curve showing two phase resistance model.

coefficients k and $\bar{k}$. The last term represents an overall film solution driving force expression where the concentration of protein at the interface C_i is replaced by C^* the concentration in the solution that would be in equilibrium with the average particle concentration $\bar{C}$ at any time. Equation 1 may be rearranged as in Treybal (1980) as follows.

The first two terms of Eq. 1 may be solved to obtain

$$\frac{\bar{C} - \bar{C}_i}{C - C_i} = -\frac{k}{\bar{k}} \qquad (2)$$

With the aid of Figure 1 which represents a typical adsorption process the individual driving forces and the overall driving force may be shown to be related. In Figure 1, P is any point along the operating line between the points $(C(o), \bar{C}(o))$ and $(C_\infty, \bar{C}_\infty)$. Note for a simple batch operation the slope of the operating line is $-V/\bar{V}$.

From the geometry of Figure 1 one may see that for any point P;

$$(C - C^*) = (C - C_i) + (C_i - C^*) \qquad (3)$$

$$= (C - C_i) + \frac{(\bar{C}_i - \bar{C})}{m'} \qquad (4)$$

where m' is slope of the chord as shown on Figure 1.

Substituting Eq. 1 into Eq. 4 one obtains

$$\frac{N}{K} = \frac{N}{k} + \frac{N}{\bar{k}m'} \qquad (5)$$

or

$$\frac{1}{K} = \frac{1}{k} + \frac{1}{\bar{k}m'} \qquad (6)$$

It can be seen that m' approximates the slope of the equilibrium line and will go from large values at the start of the process to small values as the less curved section of the equilibrium line is approached.

Values for the individual phase coefficients may be approximated by fitting the coefficients of this treatment which uses a linear driving force to more exact equations such as has been done for ion exchange by Glueckauf (1955) and Helfferich (1962) for the resin phase coefficient. The relations used in this model are:

for solution film coefficient; $k = D/\delta$ (7)

for resin phase film coefficient; $\bar{k} = \frac{\pi^2 \bar{D}}{3r_o}$ (8)

where D and $\bar{D}$ are the diffusion coefficient in the solution and resin phase respectively, δ is the effective solution film thickness and r_o is the resin particle radius.

It shoud be noted that Eq. 1 also assumes the net convective flux is zero which is another simpifying approximation.

The time rate of change of $\bar{C}$ may be related to N by

$$N = \frac{\bar{V}}{\bar{A}} \frac{d\bar{C}}{dt} = \frac{r_o}{3} \frac{d\bar{C}}{dt} = K(C - C^*) \tag{9}$$

Then using conservation; $(C - C_\infty)V = (\bar{C}_\infty - \bar{C})\bar{V}$ (10)

and

$$m'' = \frac{\bar{C}_\infty - \bar{C}}{C_\infty - C^*} \tag{11}$$

where the subscript ∞ denotes conditions at the end of the process, C and C^* may be replaced in Eq. 9 to obtain

$$\frac{d\bar{C}}{(\bar{C}_\infty - \bar{C})} = \frac{-3K}{r_o}\left[\frac{\bar{V}}{V} + \frac{1}{m''}\right] dt \tag{12}$$

Integrating Eq. 12 from time $t = 0$ to $t = \theta$ one obtains

$$ln \frac{\bar{C}_\infty - \bar{C}}{\bar{C}_\infty - \bar{C}(o)} = \frac{-3K\theta}{r_o}\left[\frac{\bar{V}}{V} + \frac{1}{m''}\right] \tag{13}$$

Defining the fractional obtainment of equilibrium to be

$$F(t) = \frac{\bar{C}(o) - \bar{C}}{\bar{C}(o) - \bar{C}_\infty} \tag{14}$$

Equation 13 may be reduced to

$$ln\,[1 - F(t)] = \frac{-3K\theta}{r_o}\left[\frac{\bar{V}}{V}\frac{1}{m''}\right] \tag{15}$$

Equation 15 is a general solution for both adsorption and desorption. It should be noted that m'' defined in Eq. 11 is identical to m' only if the slope of the operating line $V/\bar{V} = K/\bar{K}$. However in many cases (as in this study) both $V/\bar{V}$ and $K/\bar{K}$ are large and on the order of 100 and assuming $m' = m''$ is a reasonable approximation. Two important limiting cases are useful.

Solution Film Diffusion Controlling

In the case where $(1/k) \gg (1/\bar{k}m')$, Eq. 6 becomes $K = k$ and solution film controls. Equation 15 reduces to

$$ln\,[1 - F(t)] = \frac{-3k\theta}{r_o}\left[\frac{\bar{V}}{V} + \frac{1}{m''}\right] \tag{16}$$

For this to occur m' must be very large (greater than say 1000) since $\bar{k}$ will in general be much smaller than k.

If in addition $(\bar{V}/V) \gg (1/m'')$ Eq. 16 reduces to

$$ln\,[1 - F(t)] = \frac{-3k}{r_o}\frac{\bar{V}}{V}\theta = \frac{-3D}{r_o\delta}\frac{\bar{V}}{V}\theta \tag{17}$$

Since $\bar{V}/V$ is *generally* small the above requires that m'' be very large. Since m' and m'' usually are of the same magnitude, this condition will be satisfied when solution film diffusion controls (Eq. 16) and Eq. 17 may be taken as the usual equation for solution film diffusion controlling.

Particle Diffusion Controlling

In the case $(1/\bar{k}m') \gg (1/k)$ Eq. 6 becomes $K = \bar{k}m'$ and particle diffusion controls. Equation 15 reduces to

$$ln\,[1 - F(t)] = \frac{-3\bar{k}m'\theta}{r_o}\left[\frac{\bar{V}}{V} + \frac{1}{m''}\right] \tag{18}$$

For this to occur m' must be only moderately small (less than say ~ 10) since $\bar{k}$ will in general be much smaller than k.

If in addition $(1/m'') \gg (\bar{V}/V)$ Eq. 18 reduces to

$$ln\,[1 - F(t)] = \frac{-3\bar{k}\theta}{r_o} = \frac{-\pi^2\bar{D}}{r_o^2}\theta \tag{19}$$

Since $\bar{V}/V$ is generally small the above requires that $m'' \approx m'$ be only moderately small as required for Eq. 18. Thus, Eq. 19 may be taken as the usual equation for particle diffusion controlling.

For desorption, the quantity C^* used in the overall driving force (Eq. 1) may not be physically defined for a given desorption equilibrium curve. It is better in such cases to use an alternative but equivalent model using the overall driving force based on the particle phase. The analysis is the same as for the above except one starts with

$$N = k(C - C_i) = \bar{k}(C_i - \bar{C}) = \bar{K}(\bar{C}^* - \bar{C}) \tag{20}$$

and by similar geometric arguments as used before

$$\frac{1}{\bar{K}} = \frac{1}{\bar{k}} + \frac{\bar{m}'}{k} \tag{21}$$

where $\bar{m}'$ is similar to m' but defined by the slope of a chord between a point $(C, \bar{C}^*)$ and a point at the intersection of a line drawn from any operating point $(C, \bar{C})$ with a slope equal to $-k/\bar{k}$.

Defining

$$\bar{m}'' = \frac{\bar{C}_\infty - \bar{C}^*}{C_\infty - C} \tag{22}$$

analogous to Eq. 11 and solving the time rate of change of $\bar{C}$ one obtains the following equivalent expressions.

Solution Film Diffusion Controlling

for

$$\frac{\bar{m}'}{k} \gg \frac{1}{\bar{k}} \quad ln\,[1 - F(t)] = \frac{-3k\theta}{r_o\bar{m}'}\left[1 + \bar{m}''\frac{\bar{V}}{V}\right] \tag{23}$$

for

$$\bar{m}''\frac{\bar{V}}{V} \gg 1 \text{ and } \bar{m}' \approx \bar{m}'' \quad ln\,[1 - F(t)] = \frac{-3D}{r_o\delta}\frac{\bar{V}}{V}\theta \tag{24}$$

which is identical to Eq. 17.

Particle Diffusion Controlling

for

$$\frac{1}{\bar{k}} \gg \frac{\bar{m}'}{k} \quad ln\,[1 - F(t)] = \frac{-3\bar{k}\theta}{r_o}\left[1 + \bar{m}''\frac{\bar{V}}{V}\right] \tag{25}$$

for

$$\bar{m}'\frac{\bar{V}}{V} \ll 1 \quad ln\,[1 - F(t)] = \frac{-3\bar{k}}{r_o}\theta = \frac{-\pi^2\bar{D}}{r_o^2}\theta \tag{26}$$

which is identical to Eq. 19.

In particular Eq. 25 will be of use for cases where the extreme limits of particle diffusion controlling cannot be used (Eq. 26) and an etimate of $\bar{m}''$ is required. As mentioned before for certain cases of desorption C^* and hence m'' will not be defined due to the shape of the equilibrium curve so that the corresponding Eq. 18 is not well defined.

RESULTS

Equilibrium Studies

The equilibrium results are plotted on Figure 2. The following points should be noted:

(1) Equilibration with the protein typically took 12 hours or longer, although the greater part (75%) of the equilibration was over in less than 4 hours.

(2) For low ionic strength solutions (no salt added) there appears to be a region where one may have very high concentrations of protein in the ion exchanger (100–300 mg/gm) and nearly zero concentration (a few ppm) in the solution. Also for the

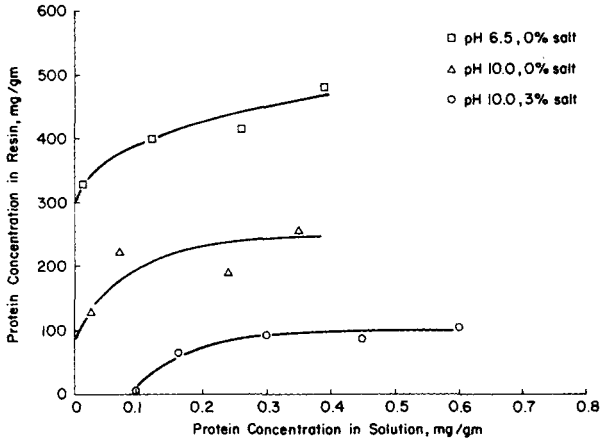

Figure 2. Equilibrium distribution of B.S.A. on DEAE "Protion."

lower pH of 6.5 which is about 1½ pH units above the isoelectric pH of B.S.A., $pI = 4.7$ (Longsworth and Jacobsen, 1949), the adsorption is considerably better than at pH of 10. This would presumably be due to the lower concentration of ionized ionic sites available at pH of 10 even though the B.S.A. itself would be more in its anionic form at pH 10.

Rate Studies

Adsorption. Results for the adsorption studies for two levels of initial protein concentration, 1.86 mg/gm soln and 3 mg/gm solution both at pH of 10 are presented in Figure 3. The results are plotted in terms of fractional attainment of equilibrium $F(t)$ versus time. For adsorption $F(t)$ is the amount of protein adsorbed at time t versus the total amount adsorbed at equilibrium.

The most striking feature of these results is the initial very fast rate period followed by a second very slow rate period. For the low and high protein concentration experiments respectively, the initial fast rate period represented 45% and 25% of the total adsorption and took place in less than 20 and 5 minutes respectively. In the slow rate period in both cases, an additional 10% of the total adsorption takes over an hour and continues for up to 1–3 days.

While one may devise many possible physical explanations and corresponding mathematical models to explain these results, the two film theory as outlined in the theory section will be used. It is of particular interest to note from the equilibrium curves (Figure 2) for adsorption at pH of 10, that for $\bar{C} < \sim 80$ mg/gm, the slope of the equilibrium line approaches infinity, while for $\bar{C} > 80$, the

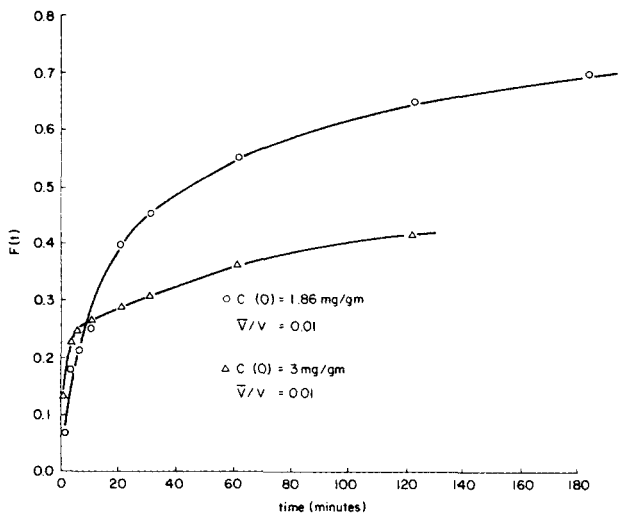

Figure 3. Rate of adsorption of B.S.A. on DEAE "Protion."

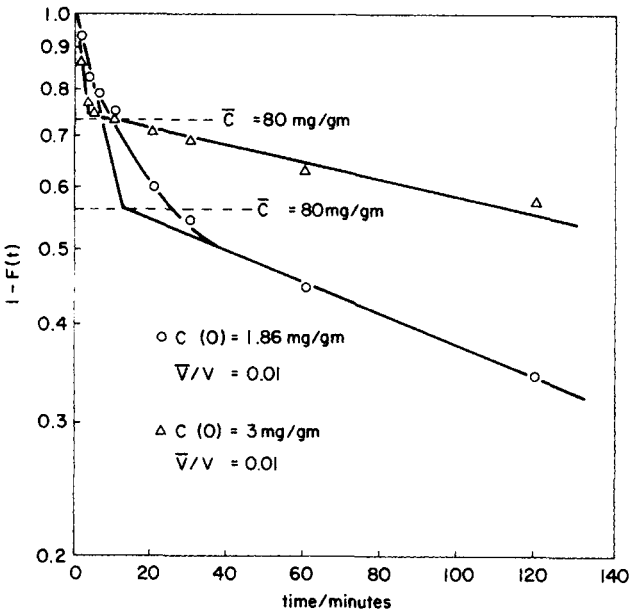

Figure 4. Plot of log [1 − $F(t)$] vs. time for adsorption of B.S.A.

slope goes to low values and finally for solution concentrations, $C > 0.1$ mg/gm (100 ppm) the slope goes to zero.

This suggests that the initial rate period will be film diffusion controlled and will last until $\bar{C} < \sim 80$ mg/gm and the final rate period will be intraparticle diffusion controlled with $m' = 0$. If we plot log $[1 - F(t)]$ versus t, take the slopes of the two rate periods and calculate the diffusion coefficients according to the two-phase diffusion equations, we may test this hypothesis. Such a plot is given in Figure 4. Note that the slope values given below are based on $ln\,[1 - F(t)]$.

Initial Rate Period:

for

$$C(o) = 3 \text{ mg/gm} \quad D = \frac{(-\text{slope})\,(r_o)(\delta)}{3(\bar{V}/V)}$$

$$= \frac{(1.3 \times 10^{-3})(0.035)(4.47 \times 10^{-4})}{3\,(0.01)}$$

$$= 7.0 \times 10^{-7} \text{ cm}^2/\text{s}$$

for

$$C(o) = 1.86 \text{ mg/gm} \quad D = \frac{(7.7 \times 10^{-4})(0.035)(4.47 \times 10^{-4})}{3\,(0.01)}$$

$$= 4.0 \times 10^{-7} \text{ cm}^2/\text{s}$$

Final Rate Period:

for

$$C(o) = 3 \text{ mg/gm} \quad \bar{D} = \frac{(-\text{slope})(r_o)^2}{(\pi^2)}$$

$$= \frac{(4.2 \times 10^{-5})(0.035)^2}{(\pi^2)}$$

$$= 5.2 \times 10^{-9} \text{ cm}^2/\text{s}$$

for

$$C(o) = 1.86 \text{ mg/gm} \quad \bar{D} = \frac{(7.2 \times 10^{-5})(0.035)^2}{(\pi^2)}$$

$$= 9.0 \times 10^{-9} \text{ cm}^2/\text{s}$$

The value of $r_o = 0.035$ applies strictly only for the OH^- form but was used throughout as an approximation. The value of film thickness, $\delta = 4.47 \times 10^{-4}$ cm was obtained in a separate experiment using Dowex 21K (a conventional anion exchange resin) in a Cl^-—OH^- exchange under identical conditions of stirring rate

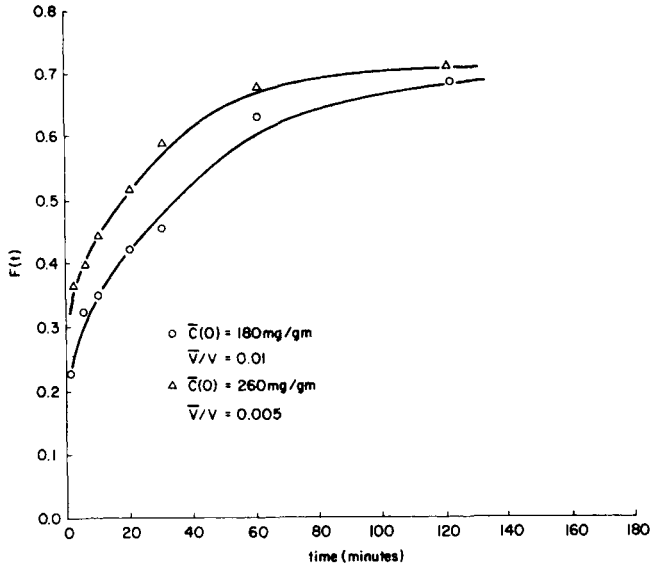

Figure 5. Rate of desorption of B.S.A. on DEAE "Protion."

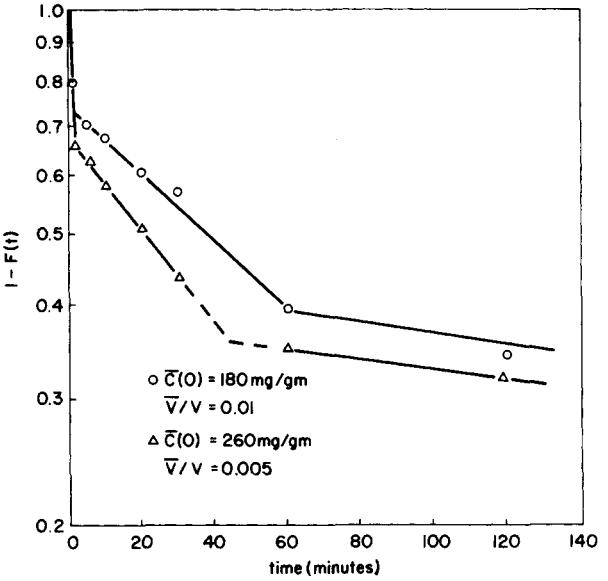

Figure 6. Plot of log [1 − $F(t)$] vs. time for Desorption of B.S.A.

and concentration, Lee (1972). The results are very encouraging with the diffusion coefficient for B.S.A. in water solution ranging from 4.0×10^{-7} to 7.0×10^{-7} cm^2/s in reasonable agreement with the value of 5.9×10^{-7} cm^2/s reported by Wagner and Scheraga (1956) for 20°C. Differences in the values obtained in these studies may be due to the different conditions used by Wagner and Scheraga. Their measurements were with 0.5 M KCl and a pH 5.1. A pH of 10 was used in these studies. The experiments of Fair, Chao and Jamieson (1978) show the effect of ionic environment on the diffusivity of B.S.A.

Also the value of $\overline{D} = 5.2 - 9.0 \times 10^{-9}$ cm^2/s is the expected order of magnitude (1/100) of diffusion in free solution, Helfferich (1961). The "change in slope" occurs around the point where $\overline{C}$ = 80 mg/gm which is the point where the equilibrium curve begins to change slope rapidly (Figure 2).

The studies of Colton, Satterfield and Lai (1979) obtained intraparticle diffusion coefficients for two proteins cytochrome C and human hemoglobin of about $1/10$ to $1/30$ of the free solution values as opposed to a ratio of $1/100$ obtained in this study. Neither the tortuosity nor exact pore size has been reported for "Protion" but estimates would indicate that the values would be of the same order of magnitude as the studies of Colton et al. (1975). However serum albumin is an ellipsoid shaped molecule of dimensions 3.8 × 15 nm so that the critical dimension is from 2–3 times larger than that of the other two proteins. Furthermore at a pH = 10 the B.S.A. would be highly ionized which is known to reduce the effective diffusivity, Helfferich (1963).

Desorption. Two of the results for desorption are given in Figure 5; one at an initial resin protein concentration of $\overline{C}(o)$ = 180 mg/gm and with $\overline{V}/V = 0.01$ (1 gm resin [dry] and 100 ml of solution) and the second at an initial protein concentration of $\overline{C}(o)$ = 260 mg/gm and with $\overline{V}/V = 0.0005$ ($1/2$ gm resin [dry] and 100 mL of solution). They were obtained using the same stirring speed of 2500 rpm and a solution that was 3% NaCl and pH 10.

A review of the equilibrium results shows that in this case m' is never very large and for the conditions of the desorption experiments we should expect particle-diffusion always to be controlling.

The results when plotted according to log [1 − $F(t)$] versus t shows, at least to a good approximation, three separate rate periods (Figure 6): an initial very fast rate period lasting for only 2 minutes but accounting for as much as 35% of the desorption: a much slower second rate period (up to 40–60 minutes) and a final very slow rate period lasting up to a full day.

The first rate period cannot be fit very well with either limiting case. It is apparently some "transient" period where a certain amount of surface protein "desorbs" very quickly and diffuses away from the surface. Assuming film diffusion controlling for both curves gives values of $D = 1.5 \times 10^{-6}$ to 3.0×10^{-6} cm^2/s. These values are an order of magnitude too high suggesting the "diffusion film" was not yet equal to the "quasi-steady state value" of 4.47×10^{-4} cm used in other experiments.

The middle rate period for both cases can best be fit using Eq. 25 for particle diffusion controlling with a value of $\overline{m}'' = 200$ estimated from the desorption equilibrium curve. This gives $\overline{D} = 7 \times 10^{-9}$ cm^2/s for the upper curve ($\overline{C}(o)$ = 180 mg/gm) and $\overline{D} = 1.5 \times 10^{-8}$ cm^2/s for the lower curve ($\overline{C}(o)$ = 260 mg/gm). These values are significantly larger than those obtained from the adsorption studies suggesting that solution film diffusion has some effect in these rate periods.

The last rate period for both cases could be fit by the particle diffusion controlling equation, Eq. 19 or 26, and gives values of $\overline{D} = 3 \times 10^{-9}$ cm^2/s for both curves in good agreement with the results from the adsorption studies.

Summarizing, these results indicate that the adsorption and desorption rates are complex for proteins involving at least two distinct rate kinetics for either process. The simple two phase resistance model can approximate the results using reasonable values or estimates of basic parameters, D, $\overline{D}$, δ and r_o. For these studies for the adsorption of B.S.A. on a DEAE cellulose based resin inspection of the equilibrium curve showed a sharp change in the curvature when the resin had adsorbed 80 mg of protein per gram of resin. The two phase resistance model shows that at this point the resistance to diffusion will change from film diffusion controlled to particle diffusion controlled. Experimental rate data showed this to be the case and the kinetics could be predicted using the value of D the free solution diffusion coefficient of B.S.A. and δ the effective film thickness measured independently for the solution diffusion controlled period. For the particle diffusion controlled kinetics a value of $\overline{D} = 1/100$ D was found and along with an average ion exchange bead radius, r_o, fit the experimental data very well. The results for the desorption kinetics were more complex showing three different rate periods. Further studies are clearly needed using different resins, proteins and experimental conditions to verify the possible generality of this very simplified model. Such studies will hopefully show ways of improving the model without greatly increasing its complexity.

NOTATION

C = protein concentration in bulk of well stirred solution at any time t, mg/gm

$\bar{C}$ = average protein concentration in resin at any time t, mg/gm
$C(o)$ = initial solution protein concentration, mg/gm
$\bar{C}(o)$ = initial resin protein concentration, mg/gm
C_∞ = final or equilibrium solution protein concentration, mg/gm
$\bar{C}_\infty$ = final or equilibrium resin protein concentration, mg/gm
C_i = protein concentration in solution at resin-solution interface, mg/gm
$\bar{C}_i$ = protein concentration in resin at resin-solution interface, mg/gm
C^* = protein concentration in the solution that would be in equilibrium with the average particle concentration $\bar{C}$ at any time, mg/gm
$\bar{C}^*$ = protein concentration in resin that would be in equilibrium with bulk solution concentration, mg/gm
D = diffusion coefficient of protein in solution, cm^2/s
$\bar{D}$ = diffusion coefficient of protein in resin, cm^2/s
$F(t)$ = fractional conversion defined by $(\bar{C}(o) - \bar{C})/(\bar{C}(o) - \bar{C}(\infty))$
k = solution side mass transfer coefficient, cm/s
$\bar{k}$ = resin side mass transfer coefficient, cm/s
K = overall mass transfer coefficient defined by Eq. 1, cm/s
$\bar{K}$ = overall mass transfer coefficient defined by Eq. 20, cm/s
m' = slope of a chord between a point defined by $(C^*, \bar{C})$ and a point at the intersection of a line drawn from any operating point $(C, \bar{C})$ with a slope equal to $-k/\bar{k}$
m'' = $(\bar{C}_\infty - \bar{C})/(C_\infty - C^*)$
$\bar{m}''$ = $(\bar{C}_\infty - \bar{C}^*)/(C_\infty - C)$
N = flux of protein, $mg/cm^2 \cdot s$
r_o = effective resin particle radius, cm
t = time, s
V = amount of solution in stirred vessel, gm
$\bar{V}$ = amount of resin in stirred vessel, gm
δ = effective solution film thickness, cm

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Studies of Mold Filling and Curing in the Reaction Injection Molding Process

A model is developed for pressure rise, extent of reaction and temperature changes during filling and curing in thin rectangular molds for the reaction injection molding (RIM) process. The predictions of the model are shown to be in good agreement with experimental results obtained for several intrumented molds using polyurethane RIM chemical systems. The relevant dimensionless groups are identified. Criteria for good mold filling are developed.

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SCOPE

The RIM process is a fairly new polymer processing method (Wood, 1974; Prepelka, 1979). Building on automotive success, it is moving into broader markets via improved systems which can give custom molding capability. The technique is still essentially an art and the trial and error approach employed in production often can not cope with variations in resin properties, complexity of molding geometries and process parameters. To seek optimum control of the molding variables, a model simulation of the process is needed.

For convenience of analysis, the RIM process can be divided into mixing, filling and curing stages as outlined by Broyer and Macosko (1976). Although typical fill times are fast, the industry trend seems to be toward larger, more complex molds and faster reactive systems. Thus, understanding the filling step is taking greater importance. Incomplete filling has been reported to give problems in complex parts (Castro et al., 1980a).

Although there have been numerous thermoplastic injection

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