

## Dialysis Continuous Process for Ammonium-Lactate Fermentation of Whey: Mathematical Model and Computer Simulation†

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A mathematical model was developed to describe a dialysis process for the continuous fermentation of whey lactose to lactic acid, with neutralization to a constant pH by ammonia. In the process, whey of a relatively high concentration is fed into the fermentor circuit at a relatively low rate so that the residual concentration of lactose is low. The fermentor effluent contains ammonium lactate, bacterial cells, and residual whey solids and could be used as a nitrogen-enriched feedstuff for ruminant animals. Only water is fed into the dialysate circuit at a relatively high rate. The dialysate effluent contains purified ammonium lactate and could be converted to lactic acid and ammonium sulfate for industry. The fermentation was specifically modeled as a set of equations representing material balances and rate relationships in the two circuits. Dialysis continuous fermentations, in general, were modeled by combining these equations and by using dimensionless parameters. The generalized model was then solved for the steady state and used to simulate the specific fermentation on a digital computer. The results showed the effects of various material and operational and kinetic parameters on the process and predicted that it could be operated efficiently.

Whey utilization is a large and worsening problem to the dairy industry. American cheesemakers annually discard about 10<sup>7</sup> metric tons of cheese whey because of its limited market value. Since about half of the solids originally in milk are left in whey, its discard is an environmental burden as well as an economic and nutrient loss.

A potential solution to this problem lies with the conversion of whey into feedstuff for ruminant animals, accomplished by the bacterial fermentation of whey lactose into lactic acid and its neutralization to a constant pH by ammonia. The primary purpose of the fermentation is to trap ammonia as ammonium lactate, thereby greatly increasing the nitrogen content of whey and decreasing the toxicity of ammonia. After condensation, the product is stable and can be used effectively as a ruminant feed supplement. The background of this development has been reviewed by Keller and Gerhardt (4) and Reddy et al. (7).

Practical usefulness is dependent on the ability to conduct the fermentation as cheaply and

efficiently as possible. It can be conducted homofermentatively without sterilization or asepsis because of the restrictive fermentation conditions of low pH (5.3 to 5.8), high concentration of undissociated acid, high temperature (44°C), and anaerobiosis. The fermentation can be conducted either in batches (7) or continuously (4), the latter process resulting in increased conversion efficiency with time.

A further increase in efficiency appeared possible by the application of dialysis (i.e., the separation of solute molecules by their unequal diffusion through a semipermeable membrane because of a concentration gradient) with the fermentation (1, 2, 8; P. Gerhardt and D. M. Gallup, U.S. Patent 3186917, June 1965). Thereby, the small molecular products are removed from the immediate environment of the bacterial cell (and ultimately from the intracellular enzyme site), thus relieving the feedback inhibition by a product that normally regulates its production. As more product is withdrawn by dialysis, more substrate is consumed and more product is made; i.e., the fermentation becomes more efficient. As the cell population attains high density, the substrate is increasingly converted into product by maintenance rather than by growth metabolism, thus also improving

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fermentation efficiency. The dialysis separation of much of the small molecular products additionally represents a purification step towards alternative uses, such as the conversion of ammonium lactate into lactic acid for use as an industrial chemical and into ammonium sulfate for use as a fertilizer (6). On the other hand, the application of dialysis increases cost, complicates operation, and eventually reduces efficiency because of membrane fouling. The potential advantages must compensate the disadvantages if a dialysis process is to be useful.

In our investigations, the goal was to assess the feasibility of applying membrane technology to the fermentation. Continuous operation appeared desirable not only for the best use of membrane technology but also because waste whey is generated and the fermentation products could be used more or less continuously. Among the several membrane techniques, dialysis appeared desirable for the initial study because of its simplicity and the existence of a theoretical basis for its application to bacterial culture (8).

In the studies reported in this article, our objective was to develop a mathematical model of a dialysis continuous process specifically for the ammonium-lactate fermentation of whey, but generalized with dimensionless parameters so that the model could be applied to other fermentations. The resulting set of equations was solved for the steady state and used to simulate the specific fermentation, on a digital computer. The experimental tests are reported in a companion article (9).

### MATHEMATICAL MODEL

**Design of fermentation system.** Figure 1 shows a schematic of a completely continuous dialysis system designed for the ammonium-lac-

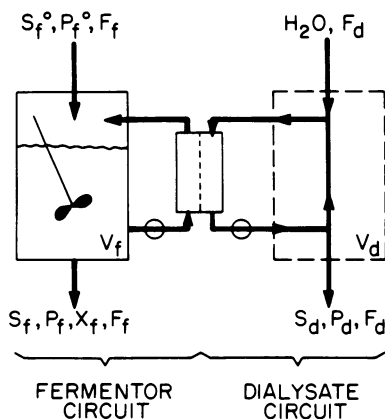


FIG. 1. Schematic of dialysis continuous fermentation system. Symbols are described in Table 1.

tate fermentation of whey. The symbols correspond to those used previously (4, 8) and are listed in Table 1. The system differs from the usual dialysis culture systems (see Fig. 8 in reference 8) following an earlier idea to dialyze a fermentation against a stream of water (see Fig. 3 in P. Gerhardt and D. M. Gallup, U.S. Patent 3186917, June 1965). This provides the greatest concentration gradient possible for dialysis and exploits the greater permeation rate of the product (lactate) rather than that of the substrate (lactose), thus maximizing efficiency of the fermentation.

The feed into the fermentor circuit contains the substrate (lactose) in relatively high concentration ( $S_f^0$ ) and is maintained at a relatively low rate of flow ( $F_f$ ) consistent with a minimal concentration of unused substrate in the fermentor effluent ( $S_f$ ). The substrate is converted essentially only to a single product (lactic acid) that remains in the fermentor effluent ( $P_f$ ). A small concentration of the product may preexist in the whey feed ( $P_f^0$ ). The liquid volume in the fermentor ( $V_f$ ) and the cell population ( $X_f$ ) are maintained at constant levels. The fermentor contents are thoroughly mixed and are continuously circulated through one side of the dialyzer. The addition of ammonia solution is not shown because it does not enter into the modeling.

The usual reservoir vessel for dialysate is eliminated. Instead, the dialysate circuit consists only of the tubing, pump, and dialysate side of the dialyzer and contains a relatively small volume ( $V_d$ ). The feed into the dialysate circuit consists only of water and is maintained at a relatively high flow rate ( $F_d$ ) consistent with a very low substrate concentration ( $S_d$ ) and a practically useful product concentration ( $P_d$ ) in the dialysate effluent.

For purposes of mathematical modeling, the assumptions are made that high mixing and circulation rates insure homogeneity throughout the system, liquid turbulence and excess membrane surface insure insignificant fouling of the dialyzer membranes for a practically useful period, bacterial metabolic rates remain constant, the volume of the dialysate circuit is negligible relative to that of the fermentor, the rate of ammonia-solution addition is negligible relative to that of substrate feed, and pressures are equalized between both circuits.

**Material-balance equations.** A set of material-balance equations was developed analogous to those in previous dialysis-culture theory (see equations 21, 22, 23, 51, and 52 in reference 8). The equations were formulated in general as follows: input + production = output + accumulation.

Equations for the substrate, product, and cell

TABLE 1. Glossary of mathematical symbols

Symbol	Description	Units
$A_m$	Area of membrane available for dialysis	cm <sup>2</sup>
$F_d$	Flow rate into and out of dialysate circuit	ml/h
$F_f$	Flow rate into and out of fermentor circuit	ml/h
$K_p$	Product inhibition constant	mg/ml
$K_s$	Michaelis-Menten saturation constant	mg/ml
$P_d$	Product concentration in dialysate circuit	mg/ml
$P_f^o$	Product concentration in fermentor feed	mg/ml
$P_f$	Product concentration in fermentor circuit	mg/ml
$P_{mp}$	Permeability of membrane to product	mg/cm <sup>2</sup> -h
$P_{ms}$	Permeability of membrane to substrate	mg/cm <sup>2</sup> -h
$r_g$	Rate of cell growth	mg/ml-h
$r_p$	Rate of product formation	mg/ml-h
$-r_s$	Rate of substrate utilization	mg/ml-h
$S_d$	Substrate concentration in dialysate circuit	mg/ml
$S_f^o$	Substrate concentration in fermentor feed	mg/ml
$S_f$	Substrate concentration in fermentor circuit	mg/ml
$t$	Time	h
$V_d$	Volume of liquid in dialysate circuit	ml
$V_f$	Volume of liquid in fermentor circuit	ml
$X_f$	Cell-mass concentration in fermentor circuit	mg/ml
$\alpha$	Substrate/cell ratio	mg/mg
$\beta$	Specific maintenance rate	h <sup>-1</sup>
$\gamma$	Product/substrate ratio	mg/mg
$\mu_m$	Maximum specific growth rate of cells	h <sup>-1</sup>
$T_f$	Cell retention time in fermentor circuit	h

mass in the fermentor circuit are as follows:

$$F_f S_f^o + V_f r_s = F_f S_f + P_{ms} A_m \cdot (S_f - S_d) + V_f \frac{dS_f}{dt} \tag{1}$$

$$\frac{F_f S_f^o}{F_f + P_{ms} A_m} + \frac{V_f}{F_f + P_{ms} A_m} r_s = - \frac{P_{ms} A_m}{F_f + P_{ms} A_m} S_d + S_f \tag{6}$$

$$F_f P_f^o + V_f r_p = F_f P_f + P_{mp} A_m \cdot (P_f - P_d) + V_f \frac{dP_f}{dt} \tag{2}$$

$$+ \left[ \frac{V_f}{F_f + P_{ms} A_m} \right] \frac{dS_f}{dt}$$

$$V_f r_g = F_f X_f + V_f \frac{dX_f}{dt} \tag{3}$$

$$\frac{F_f P_f^o}{F_f + P_{mp} A_m} + \frac{V_f}{F_f + P_{mp} A_m} r_p = - \frac{P_{mp} A_m}{F_f + P_{mp} A_m} P_d + P_f \tag{7}$$

The symbols in these and subsequent equations are described in Table 1.

For the dialysate circuit, the corresponding equations are:

$$P_{ms} A_m (S_f - S_d) = F_d S_d + V_d \frac{dS_d}{dt} \tag{4}$$

$$P_{mp} A_m (P_f - P_d) = F_d P_d + V_d \frac{dP_d}{dt} \tag{5}$$

The terms  $P_{ms} A_m (S_f - S_d)$  and  $P_{mp} A_m (P_f - P_d)$  are based on Fick's law and describe the diffusion of substrate and product, respectively, across the dialyzer membrane.

To establish maximal retention times for the substrate, product, and cells in the fermentor, equations 1, 2, and 3 can be modified as follows:

$$+ \left[ \frac{V_f}{F_f + P_{mp} A_m} \right] \frac{dP_f}{dt}$$

$$\frac{V_f}{F_f} r_g = X_f + \left[ \frac{V_f}{F_f} \right] \frac{dX_f}{dt} \tag{8}$$

Maximal retention times can be calculated from the terms within brackets in the above equations. If no bacteria existed in the fermentor, the differential equations would be linear and the actual retention times would equal the maximal retention times. From these times, the period necessary to reach a new steady state after the occurrence of a step change in an input variable can be estimated. From a change or start-up, the system will have moved 95% to a new steady state in three retention time periods,

and the system will reach steady state in a small fraction of the expected duration of continuous operation before membrane fouling becomes excessive. Consequently, the study was restricted to steady-state behavior, without integrating the equations for transient behavior.

Calculations of the terms within brackets in the above equations also show that the cell retention time is always longer than that of the substrate or product. The long cell retention time and short product retention time enable a high rate of substrate conversion in dialysis continuous fermentation.

**Rate-relationship equations.** The term for the rate of substrate utilization includes terms for both cell growth and maintenance (5):

$$-r_s = \alpha r_g + \beta X_f \quad (9)$$

The rate of product formation is proportional to that of substrate utilization (4):

$$r_p = -\gamma r_s \quad (10)$$

Although substrate limitation was not considered significant in a previous modeling of (non-dialysis) continuous whey fermentation (4), the higher substrate conversion rates resulting from dialysis may cause substrate concentrations to occur at limiting levels. Consequently, the term for cell growth rate includes terms for substrate limitation as well as product inhibition (3):

$$r_g = \mu_m \left( \frac{S_f}{K_s + S_f} \right) \left( \frac{1}{1 + P_f/K_p} \right) X_f \quad (11)$$

**Generalized model.** The above equations for rate relationships were combined with those for material balances and the variables were defined in dimensionless parameters (Table 2) to obtain a generalized model for dialysis continuous fermentation.

The resulting equations for the fermentor circuit are as follows:

$$\frac{d\bar{S}_f}{dt} = \left[ -(1 + \Pi)\bar{S}_f - \left[ \theta \left( \frac{\bar{S}_f}{\bar{K}_s + \bar{S}_f} \right) \left( \frac{1}{1 + \bar{P}_f/\bar{K}_p} \right) + \beta T_f/\alpha \right] \bar{X}_f + \Pi\bar{S}_d + 1 \right] / T_f \quad (12)$$

$$\frac{d\bar{P}_f}{dt} = \left[ -(1 + R\Pi)\bar{P}_f + \left[ \theta \left( \frac{\bar{S}_f}{\bar{K}_s + \bar{S}_f} \right) \left( \frac{1}{1 + \bar{P}_f/\bar{K}_p} \right) + \beta T_f/\alpha \right] \bar{X}_f + R\Pi\bar{P}_d + \bar{P}_f^0 \right] / T_f \quad (13)$$

$$\frac{d\bar{X}_f}{dt} = \left[ \theta \left( \frac{\bar{S}_f}{\bar{K}_s + \bar{S}_f} \right) \left( \frac{1}{1 + \bar{P}_f/\bar{K}_p} \right) - 1 \right] \bar{X}_f / T_f \quad (14)$$

where  $T_f = V_f/F_f$ .

For the dialysate circuit, the corresponding equations are:

$$\frac{d\bar{S}_d}{dt} = \left[ -(1 + \phi\Pi)\bar{S}_d + \phi\Pi\bar{S}_f \right] F_d/V_d \quad (15)$$

$$\frac{d\bar{P}_d}{dt} = \left[ -(1 + \phi R\Pi)\bar{P}_d + \phi R\Pi\bar{P}_f \right] F_d/V_d \quad (16)$$

**Generalized steady-state solution.** The time derivatives of the generalized model were set at zero to obtain a generalized solution for the steady state.

TABLE 2. Glossary of dimensionless parameters

Type	Symbol and definition	Description
Material parameters	$\bar{P}_d = P_d/\gamma S_f^0$	Product factor in dialysate circuit
	$\bar{P}_f = P_f/\gamma S_f^0$	Product factor in fermentor circuit
	$\bar{S}_d = S_d/S_f^0$	Substrate factor in dialysate circuit
	$\bar{S}_f = S_f/S_f^0$	Substrate factor in fermentor circuit
	$\bar{X}_f = \alpha X_f/S_f^0$	Cell factor in fermentor circuit
Operational parameters	$R = P_{mp}/P_{ms}$	Ratio of product/substrate membrane permeabilities
	$\Pi = P_{ms}A_m/F_f$	Membrane permeability factor
	$\phi = F_f/F_d$	Flow-rate ratio
Kinetic parameters	$\bar{K}_s = K_s/S_f^0$	Michaelis-Menten saturation factor
	$\bar{K}_p = K_p/\gamma S_f^0$	Product-inhibition factor
	$\theta = \mu_m T_f$	Time factor
Conversion-efficiency parameter	$E = 1 - \bar{S}_f - \bar{S}_d/\phi$	Fraction of substrate converted to product

Equations 15 and 16 were solved for the dimensionless substrate and product factors in the dialysate circuit ( $\bar{S}_d$  and  $\bar{P}_d$ ) in terms of  $\bar{S}_f$  and  $\bar{P}_f$ , respectively:

$$\bar{S}_d = \phi \pi_s \bar{S}_f \quad (17)$$

$$\bar{P}_d = \phi \pi_p \bar{P}_f \quad (18)$$

where  $\pi_s = \Pi / (1 + \phi \Pi)$  and  $\pi_p = R \Pi / (1 + \phi R \Pi)$ .

Equations 12 and 13 were combined to yield an equation in terms of  $\bar{S}_f$ ,  $\bar{P}_f$ ,  $\bar{S}_d$ , and  $\bar{P}_d$ . The further substitution of equations 17 and 18 resulted in an expression only in terms of  $\bar{S}_f$  and  $\bar{P}_f$ . Equation 14 was used to obtain another expression in terms of  $\bar{S}_f$  and  $\bar{P}_f$ . These two expressions were combined to obtain one (quadratic) equation for  $\bar{S}_f$  and another for  $\bar{P}_f$ :

$$\bar{S}_f^2 + \left[ K_s + \frac{\bar{K}_p(1 + \pi_p)(\theta - 1) - 1 - P_f^\circ}{1 + \pi_s} \right] \bar{S}_f \quad (19)$$

$$- \bar{K}_s \left[ \frac{\bar{K}_p(1 + \pi_p) + 1 + P_f^\circ}{1 + \pi_s} \right] = 0$$

$$\bar{P}_f = \bar{K}_p \left[ \theta \frac{\bar{S}_f}{\bar{K}_s + \bar{S}_f} - 1 \right] \quad (20)$$

Equation 19 has two real and distinct roots, one positive and one negative. The positive root is the only physically valid solution.

Equation 14 was used to eliminate the nonlinear term in equation 12, and the resulting expression was solved for  $\bar{X}_f$  as follows:

$$\bar{X}_f = \frac{1 - \bar{S}_f + \Pi(\bar{S}_d - \bar{S}_f)}{1 + \beta \Gamma_f / \alpha} \quad (21)$$

Equations 17 to 21 comprise a generalized steady-state solution for substrate, product, and cells in the fermentor and the dialysate circuits of the system. This solution was found to be stable locally.

### COMPUTER SIMULATION

The generalized mathematical model was used to simulate the dialysis continuous process for the fermentation of whey lactose into ammonium lactate. The model was programmed on a digital computer. Because the simulations were performed before actual experimental fermentations were conducted, approximations of various conditions were used as shown in Table 3, unless otherwise stated.

The dimensionless parameters that were manipulated in the simulations are included in Table 2. The time factor ( $\theta$ ), the flow-rate ratio ( $\phi$ ), and the membrane permeability factor ( $\Pi$ ) each can be adjusted experimentally. The time

factor ( $\theta$ ) was selected as the primary dimensionless variable because it included the important design parameter  $V_f/F_f$  and eliminated the need for specific values of  $\mu_m$ . The permeability ratio ( $R$ ) characterizes the molecular sensitivity of the membrane. The Michaelis-Menten saturation factor ( $\bar{K}_s$ ) and the product-inhibition factor ( $\bar{K}_p$ ) are the only parameters that are characteristic of a specific system, i.e., the ammonium-lactate fermentation of whey, but are generally relative to feed concentration. Therefore, the model could be applied to other systems without difficulty.

The simulations were made first with the two main experimental operating parameters, cell retention time in the fermentor circuit ( $\Gamma_f$ ) and flow rate into and out of the dialysate circuit ( $F_d$ ). To obtain values for  $\Gamma_f$  from the time factor ( $\theta$ ), the maximum specific growth rate ( $\mu_m$ ) was set at  $0.27 \text{ h}^{-1}$  (see Fig. 5 in reference 4). Figure 2 shows the simulated effects of changes in  $\Gamma_f$  on the concentration of substrate in the fermentor

TABLE 3. Values used in computer simulations

Symbol <sup>a</sup>	Value
$\bar{K}_p$	0.2
$\bar{K}_s$	0.001
$P_f^\circ$ (mg/ml)	0
$R$	3.0
$S_f^\circ$ (mg/ml)	230
$\gamma$	0.9
$\Pi$	10

<sup>a</sup> Symbols are described in Tables 1 and 2.

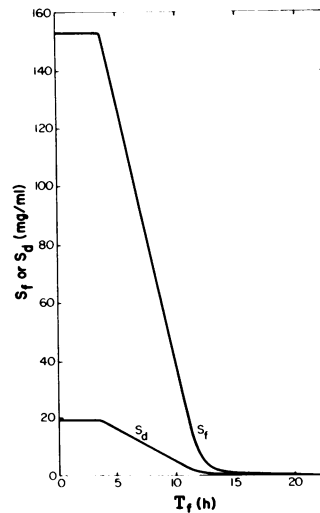


FIG. 2. Simulated effects of changes in cell retention time ( $\Gamma_f$ ) on substrate concentrations in the fermentor circuit ( $S_f$ ) and the dialysate circuit ( $S_d$ ) at constant flow rates, when  $\phi = 0.25$  and  $\Pi = 0.575$ .

tor circuit ( $S_f$ ) and the dialysate circuit ( $S_d$ ) at constant flow rates. Below a critical threshold value of  $T_f$ , the values of  $S_f$  and  $S_d$  remain constant because of cell washout. Above the threshold,  $S_f$  and  $S_d$  decrease as  $T_f$  increases. In each circuit, a plateau region is reached at which only marginal improvements occur with further increases in  $T_f$ . The simulations predicted that satisfactorily low levels of residual substrate (<5 mg/ml) could be attained by use of a reasonably short retention time in the fermentor circuit (<20 h). As expected, the general shape of the curves was the same as that for the nondialysis continuous process (see Fig. 3 in reference 4).

Figure 3 shows the simulated effects of changes in the dialysate flow rate ( $F_d$ ) on the concentration of substrate in the fermentor circuit ( $S_f$ ) and the dialysate circuit ( $S_d$ ) at constant cell retention time. Both of the dependent variables decrease as  $F_d$  increases, and satisfactorily low levels of residual substrate apparently could be attained in both circuits with a reasonably low flow rate in the dialysate circuit. In both of the simulations, the concentration of residual substrate (rather than the concentration of accumulated product) was used as a measure of fermentation conversion efficiency because of greater accuracy and precision in the experimental analysis for lactose than for lac-

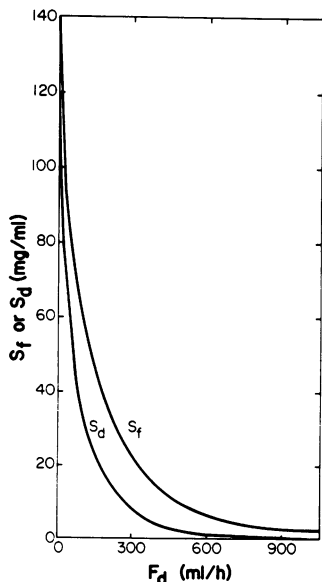


FIG. 3. Simulated effects of changes in dialysate flow rate ( $F_d$ ) on substrate concentrations in the fermentor circuit ( $S_f$ ) and the dialysate circuit ( $S_d$ ) at constant cell retention time ( $T_f = 13$  h), when  $\Pi = 0.575$ .

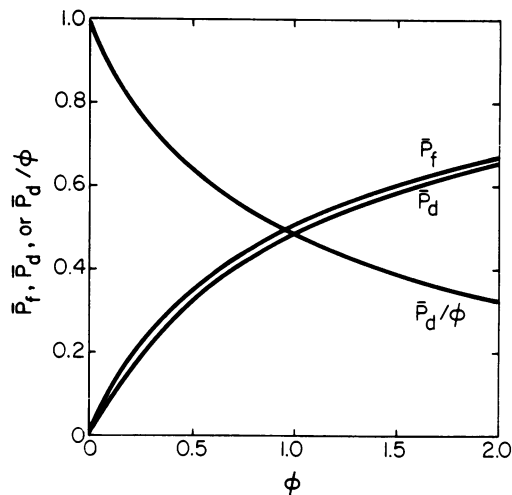


FIG. 4. Simulated effects of changes in the flow-rate ratio ( $\phi$ ) on the product factor in the fermentor circuit ( $\bar{P}_f$ ) and the dialysate circuit ( $\bar{P}_d$ ) and on the dialysate product yield ( $\bar{P}_d/\phi$ ), when  $E = 0.995$ .

tate. Furthermore, substrate use is more applicable in a generalized model because there are often several products, but there is usually only one substrate.

Generalized simulations then were made to illustrate the interrelationships of the dimensionless parameters. Figure 4 shows the simulated effects of changes in the flow-rate ratio ( $\phi$ ) on the product factors in the fermentor circuit ( $\bar{P}_f$ ) and the dialysate circuit ( $\bar{P}_d$ ) and on the dialysate product yield ( $\bar{P}_d/\phi$ ), which represents the fraction of substrate remaining as product in the dialysate effluent. As  $\phi$  increases,  $\bar{P}_f$  and  $\bar{P}_d$  increase and  $\bar{P}_d/\phi$  decreases. However, to achieve a preselected level of efficiency ( $E = 0.995$ ), the time factor ( $\theta$ ) must be adjusted for changes in  $\phi$  (Fig. 5); i.e.,  $T_f$  must be increased with increasing changes in  $\phi$  to maintain a low level of substrate in both circuits. In the ammonium-lactate fermentation, it is desirable to have high values of  $\bar{P}_f$ ,  $\bar{P}_d$ , and  $\bar{P}_d/\phi$ . Since  $\theta$  varies with  $\phi$ , a compromise probably would be made considering cost of the facility and relative marketability of the two effluents.

Figure 5 also shows the effect of changes in the product-inhibition factor ( $\bar{K}_p$ ) on the dialysis continuous fermentation. The smaller the value of  $\bar{K}_p$ , the more effective dialysis becomes.

The effect of changes in the membrane permeability factor ( $\Pi$ ) on the product factor in the fermentor circuit ( $\bar{P}_f$ ) and the dialysate circuit ( $\bar{P}_d$ ) at three permeability ratios ( $R$ ) is shown in Fig. 6. Each of the curves reaches a plateau region where a further increase in  $\Pi$  does little to decrease  $\bar{P}_f$  or increase  $\bar{P}_d$ . A high  $\Pi$  is needed

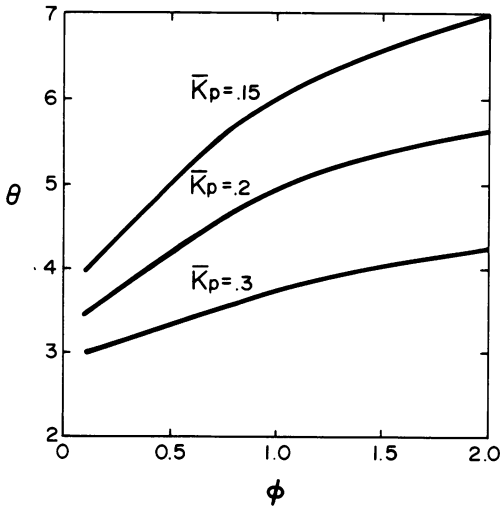


FIG. 5. Simulated effects of changes in the flow-rate ratio ( $\phi$ ) on the time factor ( $\theta$ ) at three values of the product-inhibition factor ( $\bar{K}_p$ ), when  $E = 0.995$ .

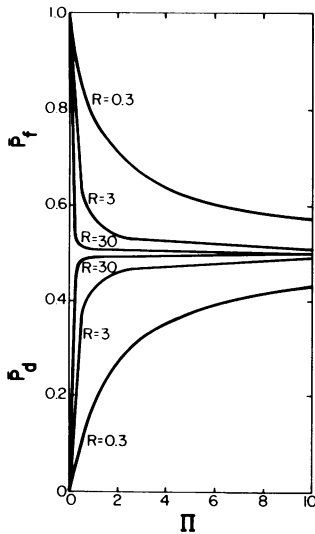


FIG. 6. Simulated effects of changes in the membrane permeability factor ( $\Pi$ ) on the product factor in the fermentor circuit ( $P_f$ ) and the dialysate circuit ( $P_d$ ) at three permeability ratios ( $R$ ), when  $\phi = 1.0$  and  $E = 0.995$ .

only when  $R$  is low, and a  $\Pi$  value greater than 2.0 probably would prove unnecessary in the ammonium-lactate fermentation.

The effect of changes in the time factor ( $\theta$ ) on the conversion efficiency parameter ( $E$ ) at three membrane permeability factors ( $\Pi$ ) is shown in Fig. 7. As  $\theta$  increases,  $E$  increases until a plateau region is reached. The relationship illustrates the tradeoff between the two parameters in designing the system.

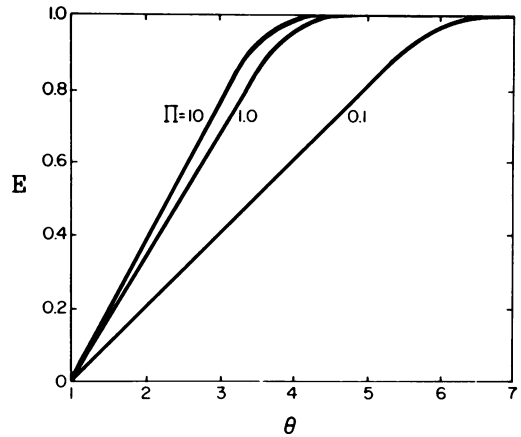


FIG. 7. Simulated effects of changes in the time factor ( $\theta$ ) on the conversion efficiency parameter ( $E$ ) at three membrane permeability factors ( $\Pi$ ), when  $\phi = 1.0$ .

### DISCUSSION

Mathematical modeling and computer simulation provide powerful tools for predicting the results to be expected in a fermentation process. Laboratory experiments then need be conducted only to validate the predictions by using a relatively limited number of changes at preselected critical points. The experimental results in turn are used to establish constants and perhaps to indicate additional terms in the mathematical equations. By this process of successive theoretical prediction and experimental validation, the model becomes increasingly accurate and useful to predict how the fermentation should be conducted for accomplishing objectives.

The foregoing generalized model and specific simulations clearly predict that a dialysis continuous process for the ammonium-lactate fermentation of whey can be operated efficiently, enabling the rapid and almost complete conversion of a high concentration of substrate into high concentrations of product and cells in one effluent and an adequate concentration of purified product in another effluent. The stimulations also project the effects of changes in various parameters on the process and the regions for experimental validation.

Although generalized, the mathematical model was developed within certain specifications related to the steady-state conversion of substrate primarily into a product that exerts feedback inhibition. Transient as well as steady-state solutions sometimes are obtained from a model by integration, but this consideration was unnecessary for the present fermentation.

Cell yield rather than product yield sometimes is more important than in the present ferment-

tation. The generalized expression for cells in the fermentor circuit (equation 21) could be simulated but would require establishing values for  $\alpha$  and  $\beta$ , which depend on the organism used. In equation 21, the cell population term ( $\bar{X}_f$ ) increases as the term  $\beta T_f/\alpha$  decreases. Thus, in situations where substrate utilization is more a function of growth ( $\alpha$ ) than of maintenance ( $\beta$ ), the cell population would increase with retention time ( $T_f$ ). Beyond a critical retention time, however, maintenance supersedes growth and then the cell population would actually decrease. In a previous mathematical model of dialysis continuous culture,  $X_f$  was predicted to increase with  $T_f$  (i.e., with decreasing dilution rate); however, the theory did not consider values of  $T_f > 10$  h (see p. 21-25 in reference 8). Such considerations and simulations of cell yield would be useful if the conversion of whey were designed to produce single-cell protein rather than ammonium lactate as the primary product or if the process were modeled to produce dairy starter cultures. The generalized nature of the model enables such alternative applications.

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