Optimal Design of an Integrated Fermentation Process for Lactic Acid Production

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In this work, we considered a multistage integrated continuous fermentation process for producing lactic acid. Each stage consists of a mixing tank, a bioreactor, a cell-recycle unit, and an extractor. A generalized mathematical model was formulated to express the integrated process. We have compared the overall productivity and conversion of the integrated process with those of two simplified processes. From the design equations, three processes have an identical overall conversion. However, the proposed process has the greatest overall productivity. A specific kinetic model for lactic production was applied to the integrated process to formulate it as a flexible or fuzzy optimization problem to find the maximum overall productivity and conversion with interval residual and supplied glucose constraints. Four optimization problems were considered to determine the optimal stages, operating conditions, and design variables. If the decision variables excluded the working volume ratio and flow rate ratio in the optimization problem, four stages were required to yield the maximum overall productivity and near complete overall conversion. However, if both operation variables were considered as the decision variables in the optimization problem, three stages were sufficient to achieve the identical overall productivity. © 2007 American Institute of Chemical Engineers *AIChE J,* 53: 449–459, 2007

Keywords: integrated process, fuzzy optimization, fermentation process, optimal design, hybrid differential evolution

Introduction

Lactic acid is a commodity chemical with a wide range of applications, mainly in the food industry, but also in the production of pharmaceuticals and cosmetics as well as in the polymer industry, where it is used for making biodegradable plastics.[1,2] The industrial production of lactic acid can be carried out by two alternative technologies: chemical synthesis from fossil fuels and biotechnological processes. Nowadays, the fermentative production of lactic acid is the world's leading technology. To increase the efficiency of the lactic acid fermentation processes, various cell culture methods have been investigated.[3–10] Achieving a high lactic acid production in a continuous bioreactor requires retaining high cell concentrations in the bioreactor and maximizing the dilution rate. As in most organic acid fermentations, end-product inhibition is a big obstacle in lactic acid fermentation. It is thus critical to remove the end product in situ and thus increase the process productivity. In the past, several processes were examined for this purpose.[9,10] The cell-recycling bioreactor coupled with membrane-filtering modules has gained considerable interest in recent years.[5,11,12] Although a high cell density can be achieved with such a cycle system, large amounts of nutrients such as carbon and nitrogen sources are wasted by the operation. Moreover, this is a water-intensive fermentation process because large amounts of wastewater must be treated in the downstream processes.

Extractive fermentation is an alternative technique used to reduce the end-product inhibition by removing the fermentation product in situ.[10,12–14] However, the toxicity of the organic

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solvent is always a problem. Kaiming et al.[13] proposed extraction fermentation using a cell-recycle system to solve this problem. There are several decision design parameters that should be considered in the design or development of an extraction fermentation system. For example, extraction fermentation performance with cell-recycle systems depends on the fermentation efficiency, the cell-recycle bleed ratio, the feed flow rate, and the fed glucose concentration. Harmand et al.[15] introduced a new result for the optimal design of two interconnected continuous stirred bioreactors in which a single reaction occurs. Wang and Lin[16] proposed a two-stage fermentation process with cell recycling including an extractor for lactic acid production. The extractive fermentation process is more efficient than two specified fermentation processes with cell recycling, as discussed in Wang and Lin.[16] In this study, the two stage extractive fermentation process is extended into a multistage integrated process to determine the optimal performance.

The integrated process is formulated as a generalized mathematical model. If some operation unit is omitted, the integrated process is reduced into two simplified processes. The first simplified process is to exclude the extractor from the integrated process. The other simplified process is a series continuous fermentation, which is equivalent to omitting both the cell-recycle unit and the extractor from the proposed process. The overall productivity and conversion of the integrated process are compared with those of the two simplified processes. In a case study, the kinetic model of the lactic fermentation process discussed in Ben Youssef et al.[17] is applied to the integrated process to obtain the maximum overall productivity and to determine the optimal stages. Enrichment is also considered in the kinetic model to enhance lactic acid production. A fuzzy decision-making method will be introduced to design the integrated fermentation process.

Mathematical Formulation

As discussed in the previous section, various cell culture methods have been investigated to increase the efficiency of lactic acid fermentation processes. Retaining high cell concentrations and decreasing end-product inhibition in a fermentation process are two main challenges to enhance the productivity of lactic acid fermentation processes. Continuous fermentation is a conventional operation to increase productivity. However, such an operation is unable to retain high cell concentrations and to achieve the maximum dilution rate because of washout. Cell-recycling fermentations can be applied to retain higher cell concentrations and avoid the washout effect. The end-product inhibition is another difficulty encountered. Typically, the accumulation of lactic acid in the fermentation broth causes an inhibitory effect on the cell culture and can eventually lead to a termination of fermentation activity. Extractive fermentation is a fermentation technique capable of handling this process limitation through integration of product generation with in situ product extraction in one single process unit.

Figure 1 shows a schematic diagram of the multistage integrated continuous process under consideration. Each stage consists of a mixing tank, a bioreactor, a cell-recycle unit, and an extractor. The sterile glucose, enrichment, and nutrient media are well stirred in the mixing tank to form a homogeneous substrate. The substrate is continuously fed into each bioreactor to produce lactic acid. Retaining a high cell concentration in each bioreactor, the broth is passed through a cell separation unit to recycle the cells back into the bioreactor. The small ratio efflux is also fed into the next bioreactor. In each cell separator, the bioactivity and residence time are negligible and the membrane filtration is perfect; thus each filtrate is cell free. The filtrate from each cell separator is transferred to an extractor in which lactic acid is extracted. A biocompatible solvent, such as a tertiary amine and oleyl alcohol,[9,14] is added into the extractor to extract lactic acid. The solvent should be biocompatible, inert to the reaction, stable under the liquid phase reaction conditions, easy to separate from lactic acid, and able to induce phase splitting. The raffinate phase in the extractor containing lactic acid, solvent, and some unconverted substrate is also transferred to the next bioreactor. Fresh substrate is added to each bioreactor.

The dynamic material balances for the biomass, glucose, product, and enrichment around the first stage, as shown in a dashed box of Figure 1, are expressed as follows:

$$\frac{dx_1}{dt} = -b_1 D_1 x_1 + r_{x_1} \tag{1}$$

$$\frac{ds_1}{dt} = D_1\left(\delta_1 s_{f_1} - s_1\right) - r_{s_1} \tag{2}$$

$$\frac{dp_1}{dt} = D_1\left(\gamma_1 p_{f_1} - p_1\right) + r_{p_1} \tag{3}$$

$$\frac{dg_1}{dt} = D_1(1 - \delta_1 - \gamma_1) g_{f_1} - D_1 g_1 \tag{4}$$

Similarly, the material balance equations around the nth stage are described as follows:

$$\frac{dx_n}{dt} = \frac{D_1}{\alpha_n}\left[b_{n-1}\left(\sum_{i=1}^{n-1}\beta_i\right)x_{n-1} - b_n\left(\sum_{i=1}^{n}\beta_i\right)x_n\right] + r_{x_n} \tag{5}$$

$$\frac{ds_n}{dt} = \frac{D_1}{\alpha_n}\left[\delta_n\beta_n s_{f_n} + \left(\sum_{i=1}^{n-1}\beta_i\right)s_{n-1} - \left(\sum_{i=1}^{n}\beta_i\right)s_n\right] - r_{s_n} \tag{6}$$

$$\frac{dp_n}{dt} = \frac{D_1}{\alpha_n}\left[\gamma_n\beta_n p_{f_n} + \left(\sum_{i=1}^{n-1}\beta_i\right)\frac{1 + b_{n-1}E_{n-1}}{1 + E_{n-1}}p_{n-1} - \left(\sum_{i=1}^{n}\beta_i\right)p_n\right] + r_{p_n} \tag{7}$$

$$\frac{dg_n}{dt} = \frac{D_1}{\alpha_n}\left[(1 - \delta_n - \gamma_n)\beta_n g_{f_n} + \left(\sum_{i=1}^{n-1}\beta_i\right)g_{n-1} - \left(\sum_{i=1}^{n}\beta_i\right)g_n\right] \tag{8}$$

where x_n, s_n, p_n, and g_n are concentrations of the cell, glucose, lactic acid, and enrichment at the nth stage, respectively. In this work, the rate equations r_{x_n}, r_{s_n}, and r_{p_n}, for biomass, glucose, and product, are considered general formulas so that the design approach for the integrated process is not only suited for the lactic acid production, but can also be applied to any fermentation processes described by unstructured models. The rate equations r_{x_n}, r_{s_n}, and r_{p_n}, for cell, glucose, and lactic acid

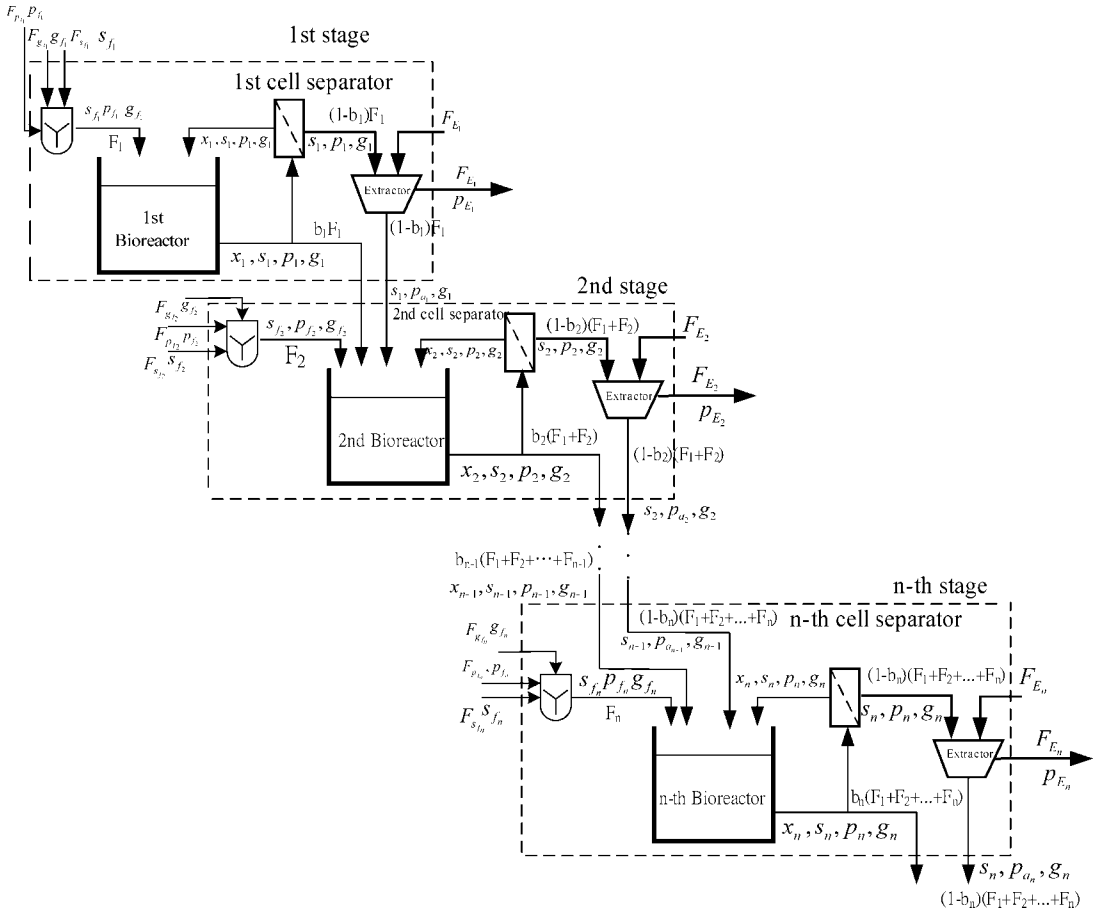

Figure 1. A multistage integrated fermentation process.

Each stage consists of a mixing tank, a bioreactor, a cell-recycle unit, and an extractor (Process A).

are in terms of the used microbial species and are calculated at the nth stage. The enrichment is used to enhance the reaction rate so its effect should be included in each rate equation. The operation parameters in Eqs. 1 to 8 are defined as follows: The dilution rate is denoted as $D_1 = F_1/V_1$, b_n is the bleed ratio at the nth stage, $\alpha_n = V_n/V_1$ is the volume ratio of the nth bioreactor to the first bioreactor, $\beta_n = F_n/F_1$ is the ratio of the overall feed flow rate at the nth stage to that at the first stage, $\delta_n = F_{s_{fn}}/F_1$ is the ratio of the glucose feed flow rate at the nth stage to the overall feed flow rate at the first stage, and $\gamma_n = F_{p_{fn}}/F_1$ is the ratio of the product feed flow rate at the nth stage to the overall feed flow rate at the first stage, which is considered zero in the case study. These parameters are considered the operation variables in the optimal design problem. By the definition, both α_1 and β_1 are equal to one. For each stage, the filtrate from the cell separator contacts the solvent in the extractor to remove the product so that the material balance for the product includes the extraction coefficient E_{n-1} at the previous stage.

The filtrate from the cell separator contacts the solvent in the extractor to remove lactic acid. The material balance for the lactic acid around each extractor can be written as

$$(1-b_n)D_1 p_n \sum_{i=1}^{n} \beta_i - D_{E_n} p_{E_n} - (1-b_n) \times D_1 \frac{p_n}{1+E_n} \sum_{i=1}^{n} \beta_i = 0 \quad (9)$$

where the solvent dilution rate is defined as $D_{E_n} = F_{E_n}/V_1$, and F_{E_n} is the solvent flow rate at the nth stage. The extraction efficiency E_n for each stage is defined as

$$E_n = \frac{p_n}{p_{a_n}} - 1 = K\frac{D_{E_n}}{(1-b_n)D_n} \quad (10)$$

where K is the distribution coefficient.

Process Performance

To design an integrated chemical process, the first decisions are those that lead to the choice of reactor. These decisions are among the most important in the whole chemical process design.[18] Good bioreactor performance is of paramount importance in determining the economic viability of the overall design. To evaluate performance of the integrated process, we define the lactic acid productivity for each stage and for the overall process as

Productivity
= (net of lactic acid production rate)/working volume
= $\{\Sigma_{\text{efflux}}(\text{flow rate} \times \text{lactic acid concentration})_{\text{efflux}} - \Sigma_{\text{influx}}(\text{flow rate} \times \text{lactic acid concentration})_{\text{influx}}\}$/working volume

The first term in the numerator is the sum of the efflux of lactic acid produced. The second term is the sum of the influx of

lactic acid from the previous stage. The conversion of glucose is another criterion for bioreactor performance analysis and is defined as

$$\text{Conversion} = (\text{glucose consumed in the process})/(\text{glucose fed to the process})$$

According to the definitions, the lactic acid productivity (π_1^A) and glucose conversion (χ_1^A) for the first stage are therefore expressed as

$$\pi_1^A = \frac{1}{V_1}\left[F_{E_1}p_{E_1} + b_1F_1p_1 + (1-b_1)F_1p_{a_1} - F_{p_{f_1}}p_{f_1}\right] \quad (11)$$

$$\chi_1^A = 1 - \frac{s_1}{\delta_1 s_{f_1}} \quad (12)$$

The lactic acid in the first bleed is accounted for as being recoverable as the product. By substituting Eqs. 9 and 10 into Eq. 11, the productivity for the first stage is therefore expressed as

$$\pi_1^A = D_1(p_1 - \gamma_1 p_{f_1}) \quad (13)$$

Similarly, the lactic acid productivity and glucose conversion for the nth stage are respectively defined as

$$\pi_n^A = \frac{D_1}{\alpha_n}\left[\left(\sum_{i=1}^{n}\beta_i\right)p_n - \left(\sum_{i=1}^{n-1}\beta_i\right)\frac{1+b_{n-1}E_{n-1}}{1+E_{n-1}}p_{n-1} - \gamma_n\beta_n p_{f_n}\right] \quad (14)$$

$$\chi_n^A = 1 - \frac{\left[\sum_{i=1}^{n}\beta_i\right]s_n}{\delta_n\beta_n s_{f_n} + \left[\sum_{i=1}^{n-1}\beta_i\right]s_{n-1}} \quad (15)$$

Note that some of the lactic acid is removed from the extractor so that the productivity for the nth stage includes the extraction efficiency.

The overall lactic acid productivity and overall glucose conversion are respectively defined as follows

$$\pi^A = \frac{\sum_{i=1}^{n}F_{E_i}p_{E_i} + b_np_n\sum_{i=1}^{n}F_i + (1-b_n)p_n\sum_{i=1}^{n}F_i - \sum_{i=1}^{n}F_{p_{f_i}}p_{f_i}}{\sum_{i=1}^{n}V_i} \quad (16)$$

$$\chi^A = \frac{F_l\sum_{i=1}^{n}\beta_i\delta_i s_{f_i} - F_l\sum_{i=1}^{n}\beta_i s_n}{F_l\sum_{i=1}^{n}\beta_i\delta_i s_{f_i}} \quad (17)$$

By substituting Eqs. 9 and 10 into Eq. 16, the overall productivity is therefore

$$\pi^A = \frac{D_1}{\sum_{i=1}^{n}\alpha_i}\left\{\left(\sum_{i=1}^{n}\beta_i\right)p_n + \sum_{i=1}^{n}(1-b_i)\frac{E_ip_i}{1+E_i} \times \left(\sum_{j=1}^{i}\beta_j\right) - \sum_{i=1}^{n}\gamma_i\beta_i p_{f_1}\right\} \quad (18)$$

We also obtain the overall glucose conversion as

$$\chi^A = 1 - \frac{\left(\sum_{i=1}^{n}\beta_i\right)s_n}{\sum_{i=1}^{n}(\delta_i\beta_i s_{f_i})} \quad (19)$$

The integrated process can be reduced into two simplified processes. Figure 2 shows the first simplified fermentation process in which each stage consists of a mixing tank, a bioreactor, and a cell-recycle unit. Figure 3 shows the second simplified process in which each stage consists of a mixing tank and a bioreactor. Material balances, productivities, and conversions for the three processes are listed in Table 1. In this table, Figure 1 refers to Process A, Figure 2 to Process B, and Figure 3 to Process C.

We assume that the three processes operate at the same operation conditions to compare the overall lactic acid productivity and overall glucose conversion for these processes. From Table 1, we obtain the difference of the overall productivity for process A and B as follows:

$$\pi^A - \pi^B = \frac{D_1}{\sum_{i=1}^{n}\alpha_i}\left\{\sum_{i=1}^{n}(1-b_i)\frac{E_ip_i}{1+E_i}\left(\sum_{j=1}^{i}\beta_j\right)\right\} \geq 0 \quad (20)$$

This indicates that the overall lactic acid productivity of Process A is always greater than that of Process B because an extractor is included in process A. However, the overall conversions for both processes are identical, that is, $\chi^A = \chi^B$. Moreover, the difference of the overall productivity between Process A and Process C is identical to that of Processes A and B. We conclude that $\pi^A > \pi^B = \pi^C$ and $\chi^A = \chi^B = \chi^C$. If all b_i values are equal to one, then Process A is reduced to Process C. This situation means that $\pi^A = \pi^B = \pi^C$.

Trade-Off Design

Multiobjective optimization

The aim of the optimization problem is to simultaneously maximize the overall lactic acid productivity and the overall glucose conversion. This problem becomes a multiobjective optimization problem (MOOP) expressed as follows:

$$\max_{\mathbf{z}} f_1 = \pi^A \quad (21)$$

$$\max_{\mathbf{z}} f_2 = \chi^A \quad (22)$$

where the operation variables $\mathbf{z}$ consist of the dilution rate D_1, the feed glucose concentrations s_{f_i}, the bleed ratio b_j, the bioreactor volume ratio α_j, and the flow rate ratio β_j for each stage. These operation variables are restricted within physically realistic boundaries.

Good bioreactor performance is of paramount importance in determining the economic viability of the overall design and is fundamentally important to the environmental impact of the integrated process. In addition to the desired lactic acid production, some glucose may be incompletely exhausted in the bioreactor. The residual glucose not only leads to a loss of revenue but can also create environmental problems. The best solution

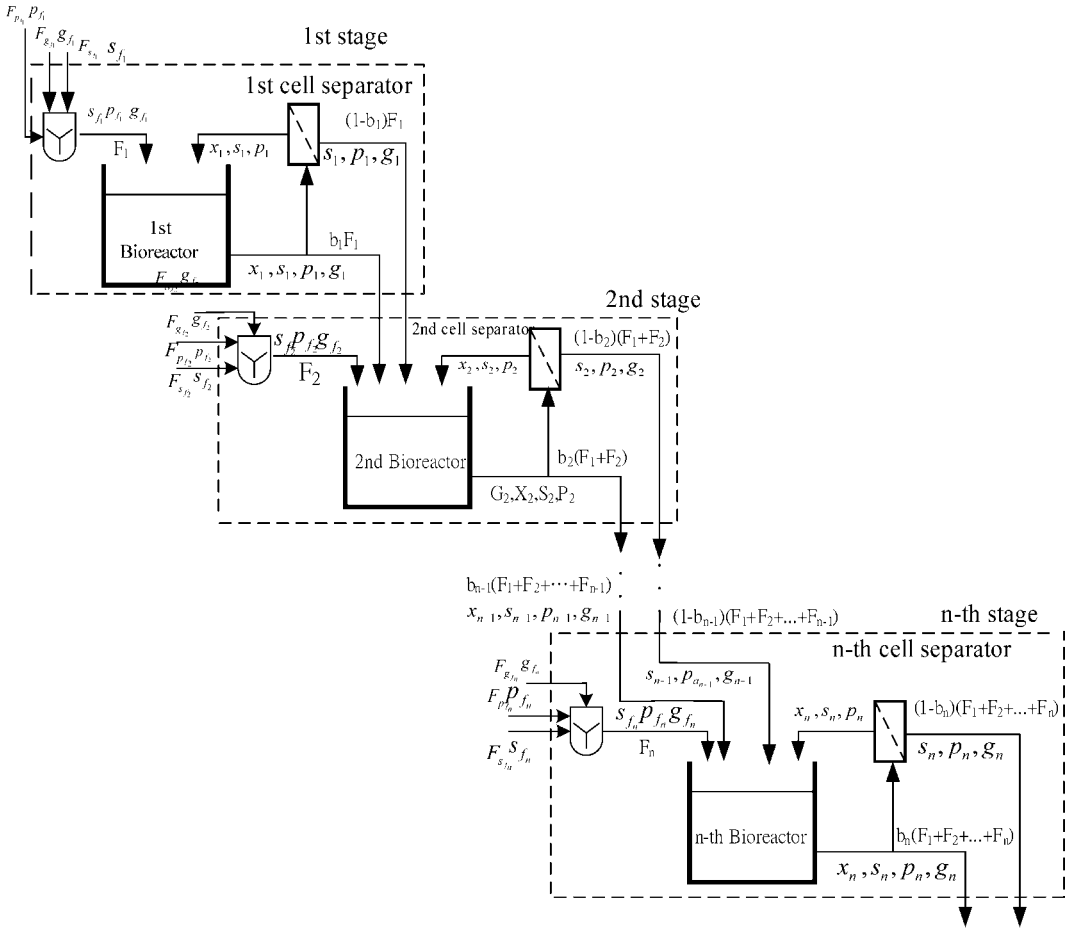

Figure 2. A simplified fermentation process.

Each stage consists of a mixing tank, a bioreactor, and a cell-recycle unit (Process B).

to environmental problems is not to use elaborate treatment methods, but to not produce waste in the first place. In this study, two additional inequality constraints are considered as the boundaries in the optimization problem. To reduce the operational cost and environmental impact, the residual glucose in the final stage is restricted by

$$s_n \leq s_r^U \tag{23}$$

where s_r^U is the crisp boundary for the desired residual glucose assigned by the decision maker (DM). The total amount of glucose fed into the process is restricted by

$$\frac{D_1\left(\sum_{i=1}^{n}\beta_i s_{fi}\right)}{\left(\sum_{i=1}^{n}\alpha_i\right)} \leq s_t^U \tag{24}$$

where s_t^U is the crisp boundary for the feed glucose flux assigned by the DM. This boundary is equivalent to the total amount of glucose supplied into the process, which is one of the investment costs.

In the real world, much of the MOOP takes place in an environment in which the DM has the preferred goals and boundaries in advance. Such a preference design problem is a research branch of decision-making problems. Two requirements must be satisfied in the decision-making problem. The first requirement is to solve the MOOP to obtain the optimal operation variables and the associated optimal objective function values and constraints. The second requirement is to check whether each optimal objective function value and constraint satisfies the preassigned goal and boundaries. If any optimal objective function value does not satisfy the goals or any constraints are violated, then the DM must make trade-offs with some goals and boundaries and repeat the problem to obtain a satisfactory solution. The weighted-sum method is one of the most commonly used techniques for MOOP to obtain a Pareto optimal solution. However, the weighted-sum method is not a preference technique, so the method is unable to handle the DM's preferred goals and boundaries. Some preference techniques, such as nonlinear goal programming, compromise programming, and surrogate worth trade-off methods, can be used to solve the decision-making problem. Such methods, however, although able to solve the problem with preferred goals, are not suited for interval constraint boundaries. In this study, we will apply a fuzzy goal attainment method to overcome such drawbacks.

Flexible goal attainment approach

In the preference design problem, the DM usually assigns an interval goal, rather than a rigid value. Here, we consider the interval goal for the productivity $[\pi^L, \pi^U]$ and for conversion

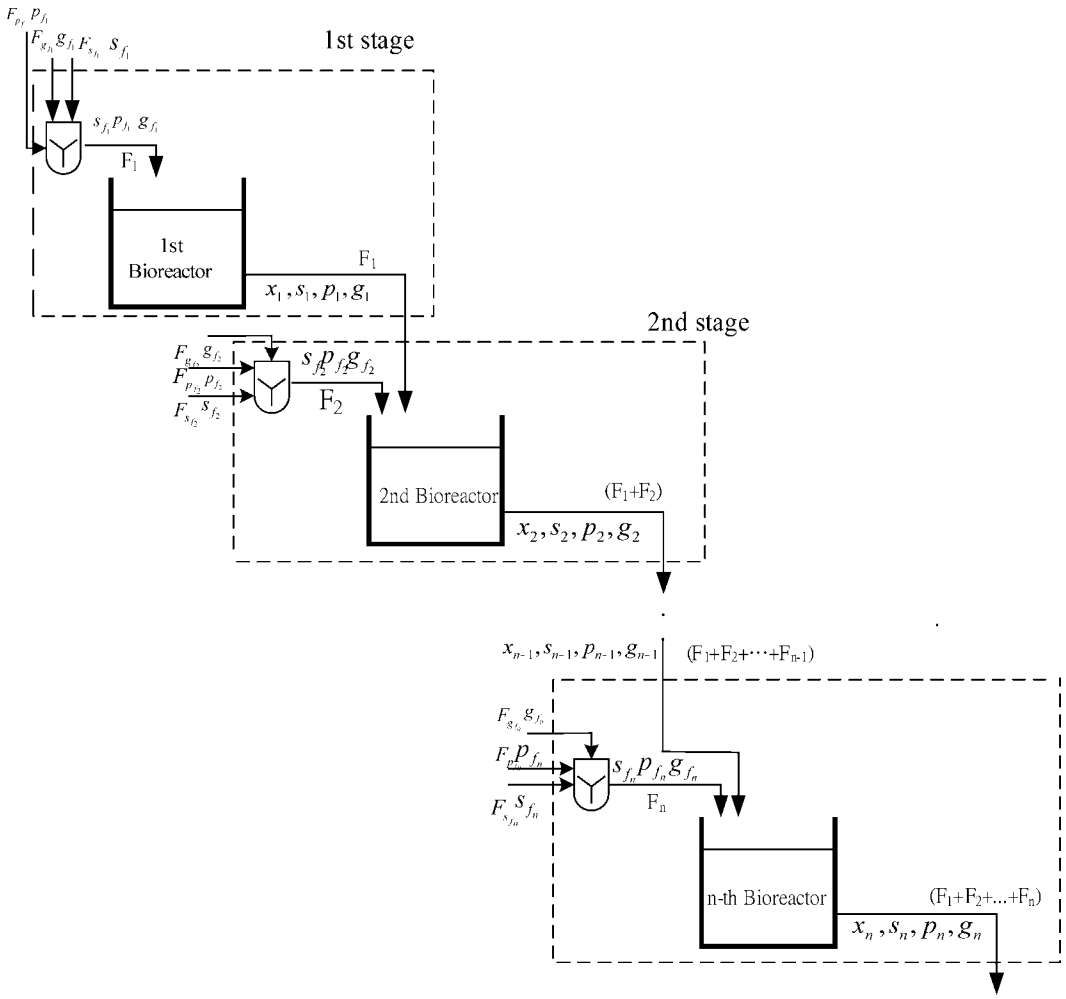

Figure 3. A continuous fermentation process.

Each stage consists of a mixing tank and a bioreactor (Process C).

$[\chi^L, \chi^U]$, respectively. The preference goal problem is therefore expressed as

$$\widetilde{\max_{\mathbf{z}}} f_1 = \pi^A \in \left[\pi^L, \pi^U\right] = \left[f_1^L,\ f_1^U\right] \tag{25}$$

$$\widetilde{\max_{\mathbf{z}}} f_2 = \chi^A \in \left[\chi^L, \chi^U\right] = \left[f_2^L,\ f_2^U\right] \tag{26}$$

This multiobjective optimization problem is referred to as a flexible or fuzzy optimization problem and indicates that the optimal solutions need to be included in the preference intervals. The interval or fuzzy goal for each objective function can be quantified by eliciting membership functions from the DM. In maximization, a fuzzy goal stated by the DM may be to achieve a result "substantially greater than or equal to some interval," and the DM is asked to determine the subjective membership function, which is a strictly monotonically increasing function with respect to f_k in the following way:

$$\eta_k(f_k) = \begin{cases} 0; & f_k \le f_k^L, \quad k = 1, 2 \\ d_k;\ f_k^L \le f_k \le f_k^U \\ 1; & f_k^U \le f_k \end{cases} \tag{27}$$

where f_k^L or f_k^U represents the value of f_k such that the grade of the membership function $\eta_k(f_k)$ is 0 or 1 and the grades of the membership for the intermediate function values are expressed by a strictly monotonically increasing function d_k with respect to f_k.

In this study, we also consider the interval inequality constraints as follows:

$$f_3 = s_n \preceq \left[s_r^L, s_r^U\right] = \left[f_3^L, f_3^U\right] \tag{28}$$

$$f_4 = \frac{D_1\left(\sum_{i=1}^n \beta_i s_{fi}\right)}{\left(\sum_{i=1}^n \alpha_i\right)} \preceq \left[s_t^L, s_t^U\right] = \left[f_4^L,\ f_4^U\right] \tag{29}$$

where the symbol "$\preceq$" denotes a relaxed or fuzzy version of the ordinary inequality "$\le$" and $[s_r^L,\ s_r^U]$ and $[s_t^L,\ s_t^U]$ are the interval boundaries for the residual and supplied glucose. The first fuzzy inequality constraint means that the DM is completely acceptable if the residual glucose in the final stage is less than s_r^L. However, the DM is absolutely unacceptable if s_n is greater than s_r^U. When s_n is between $[s_r^L,\ s_r^U]$, this implies that the DM is satisfactory to some degree. Similarly the sec-

Table 1. Material Balances, Productivities, and Conversions for Each Process*

Item	Process A	Process B	Process C
Material balances at the 1st stage	Eqs. 1–4	As same as Eqs. 1–4	The material balance equations for glucose, lactic acid and enrichment are identical to Eqs. 2, 3 and 4. The governed equation for the cell is as follows: $$\frac{dx_1}{dt} = -D_1 x_1 + r_{x_1}$$
Material balances at the nth stage	Eqs. 5–8	The material balance equations for cell, glucose and enrichment are identical to Eqs. 5, 6 and 8. The govern equation of lactic acid is as follows: $$\frac{dp_n}{dt} = \frac{D_1}{\alpha_n}\left[\gamma_n\beta_n p_{f_n} + \left(\sum_{i=1}^{n-1}\beta_i\right)p_{n-1} - \left(\sum_{i=1}^{n}\beta_i\right)p_n\right] + r_{p_n}$$	The material balance equations for glucose and enrichment are identical to Eqs. (6) and (8). The governed equation for the cell and lactic acid as follows: $$\frac{dx_n}{dt} = \frac{D_1}{\alpha_n}\left[\left(\sum_{i=1}^{n-1}\beta_i\right)x_{n-1} - \left(\sum_{i=1}^{n}\beta_i\right)x_n\right] + r_{x_n}$$ $$\frac{dp_n}{dt} = \frac{D_1}{\alpha_n}\left[\gamma_n\beta_n p_{f_n} + \left(\sum_{i=1}^{n-1}\beta_i\right)p_{n-1} - \left(\sum_{i=1}^{n}\beta_i\right)p_n\right] + r_{p_n}$$
Productivity for the 1st stage	Eq. 13	As same as Eq. 13	As same as Eq. 13
Productivity for the nth stage	Eq. 14	$$\pi_n^B = \frac{D_1}{\alpha_n}\left[\left(\sum_{i=1}^{n}\beta_i\right)p_n - \left(\sum_{i=1}^{n-1}\beta_i\right)p_{n-1} - \gamma_n\beta_n p_{f_n}\right]$$	$$\pi_n^C = \frac{D_1}{\alpha_n}\left[\left(\sum_{i=1}^{n}\beta_i\right)p_n - \left(\sum_{i=1}^{n-1}\beta_i\right)p_{n-1} - \gamma_n\beta_n p_{f_n}\right]$$
Overall productivity	Eq. 18	$$\pi^B = \frac{D_1}{\sum_{i=1}^{n}\alpha_i}\left\{\left(\sum_{i=1}^{n}\beta_i\right)p_n - \sum_{i=1}^{n}\gamma_i\beta_i p_{f_i}\right\}$$	$$\pi^C = \frac{D_1}{\sum_{i=1}^{n}\alpha_i}\left\{\left(\sum_{i=1}^{n}\beta_i\right)p_n - \sum_{i=1}^{n}\gamma_i\beta_i p_{f_i}\right\}$$
Glucose conversion for the 1st stage	Eq. 12	As same as Eq. 12	As same as Eq. 12
Glucose conversion for the nth stage	Eq. 15	As same as Eq. 15	As same as Eq. 15
Overall conversion	Eq. 19	As same as Eq. 19	As same as Eq. 19

*Process A refers to the process shown in Figure 1, Process B to Figure 2, and Process C to Figure 3.

Table 2. Best Results for the Integrated Process with Various Stages and Cases*

No. of Stages	Case	f_1^* (kg / m^3 h)	f_2^*	f_3^* (kg / m^3)	f_4^* (kg / m^3 h)	η_1^*	η_2^*	η_3^*	η_4^*	D_1^* (1 / h)
2	1	23.7900	0.9039	0.50	29.8391	0.0	0.0	0.0	1.0	6.0
	2	35.6850	0.9139	0.50	44.2730	0.0	0.0	0.0	1.0	6.0
	3	31.7200	0.9100	0.50	39.5221	0.0	0.0	0.0	1.0	6.0
	4	47.5801	0.9266	0.50	58.2193	0.0	0.0	0.0	1.0	6.0
3	1	76.2851	0.9944	0.50	86.9798	0.0	0.9304	0.0	0.9282	6.0
	2	88.6578	0.9923	0.4423	101.2967	0.2126	0.9032	0.2126	0.7520	6.0
	3	101.6475	0.9737	0.3557	118.3526	0.4791	0.5980	0.4791	0.4791	6.0
	4	103.0728	0.9994	0.3461	116.9272	0.5050	0.9934	0.5053	0.5050	5.9898
4	1	103.0978	0.9999	0.3322	116.9022	0.5055	0.9986	0.5420	0.5055	5.9945
	2	103.1008	0.9999	0.3458	116.8993	0.5055	0.9932	0.5062	0.5055	6.0000
	3	103.0994	0.9992	0.1544	116.9006	0.5055	0.9990	0.9152	0.5055	5.6796
	4	103.0837	0.9996	0.1778	116.9163	0.5052	0.9957	0.8750	0.5052	5.9984
7	1	103.1037	0.9999	0.3455	116.8962	0.5056	0.9997	0.5069	0.5056	2.3465
	2	103.1067	0.9999	0.1045	116.8934	0.5056	0.9997	0.9934	0.5056	4.0715
	3	103.0997	0.9999	0.0248	116.9004	0.5055	0.9988	1.0	0.5055	5.1054
	4	103.1074	0.9999	0.2164	116.8925	0.5057	0.9994	0.8035	0.5057	1.6836

*The preference interval boundaries for the objective function are assigned as $[f_1^L, f_1^U] = [80, 140]$, $[f_2^L, f_2^U] = [0.95, 1.0]$, and interval boundary for constraints as $[f_3^L, f_3^U] = [0.1, 0.5]$, $[f_4^L, f_4^U] = [80, 140]$.

ond fuzzy inequality constraint means that the DM is completely acceptable if the total amount of supplied glucose is less than s_t^L. However, the DM is entirely unacceptable if f_4 is greater than s_t^U. When f_4 is between $[s_t^L, s_t^U]$, this implies that the DM is satisfactory to some degree. For treating fuzzy inequality constraints, the membership function is defined as

$$\eta_k(f_k) = \begin{cases} 1, & f_k \le f_k^L, \quad k = 3,4 \\ d_k', & f_k^L \le f_k \le f_k^U \\ 0; & f_k \ge f_k^U \end{cases} \tag{30}$$

where f_k^L or f_k^U represents the value of f_k such that the grade of the membership function $\eta_k(f_k)$ is 1 or 0 and the grades of the membership for the intermediate function values are expressed by a strictly monotonically decreasing function d_k' with respect to f_k.

Having determined the membership functions from the DM for each objective function and constraint, these membership function problems can be expressed as an aggregation function. Several aggregation functions were introduced in the textbook by Sakawa.[19] The fuzzy multiobjective optimization problem is therefore converted into a min-max problem. To circumvent the need to perform the Pareto optimality test in the min-max problem, it is reasonable to use the augmented min-max problem instead of the min-max problem.[19] In the augmented min-max method, the DM first determines the membership functions as expressed in Eqs. 27 and 30. After determining the membership function for each objective function and constraint, the DM is then asked to specify the reference membership level $\bar{\eta}_k$ for each membership function. We then solve the following flexible or fuzzy goal attainment problem to obtain a Pareto optimal solution:

$$\min_{\mathbf{z}} \eta_D = \min_{\mathbf{z}} \left[\max_{k=1,2,3,4} \{\bar{\eta}_k - \eta_k(f_k)\} + \rho \sum_{k=1}^{4} |\bar{\eta}_k - \eta_k(f_k)| \right] \tag{31}$$

where ρ is a sufficiently small positive scalar. The difference between the reference membership level $\bar{\eta}_k$ and the corresponding membership function value $\eta_k(f_k)$ is used as an index to indicate how each objective function and inequality constraint is close to the DM's preference level. The summation term in Eq. 31 is used to avoid inspecting a unique test for Pareto optimality. The Pareto optimal solution depends on the DM's reference membership levels $\bar{\eta}_k$ ($k = 1, \ldots, 4$), as observed from the problem expressed in Eq. 31. In the min-max sense, the Pareto solutions nearest to the requirement or better than the reference membership levels are attainable.

Results and Discussion

In the case study, the rate equations for biomass, glucose, and lactic acid, and their corresponding kinetic data are cited from Ben Youssef et al.[17] The hybrid differential evolution (HDE) algorithm is applied to solve the flexible or fuzzy goal attainment problem to obtain a Pareto optimal solution. The HDE algorithm has succeeded in application to crisp and fuzzy optimization problems.[16,20–22] The computational algorithm was discussed in Chiou and Wang[23] in detail. To solve the fuzzy goal attainment problem, we used the exponential membership function to judge the fuzzy preference for each of the objective functions and inequality constraints. For each run, the DM assigns the preference interval boundaries for each objective function and inequality constraints as listed in Table 2. A determinant in fuzzy optimization problems to find a feasible solution is to assign the realizable preference interval boundaries. Such preference interval boundaries are chosen in terms of the DM's experience to the process. Although the assigned values are too strict, we are unable to find a solution to satisfy all objective functions and constraints. As a result, at least one of the membership functions should be zero so the optimal decision η_D^* is equal to one. Alternative preference interval boundaries have to be assigned to find a satisfied decision. An interactive procedure[16,19] can be applied to overcome such a drawback. Each single crisp optimization problem is first solved. Such optimal solutions are then served as the reference values to assign the preference interval boundaries for the fuzzy optimization problem.

In the example, the reference membership levels, $\bar{\eta}_k$, were set to one for each objective function and inequality constraint.

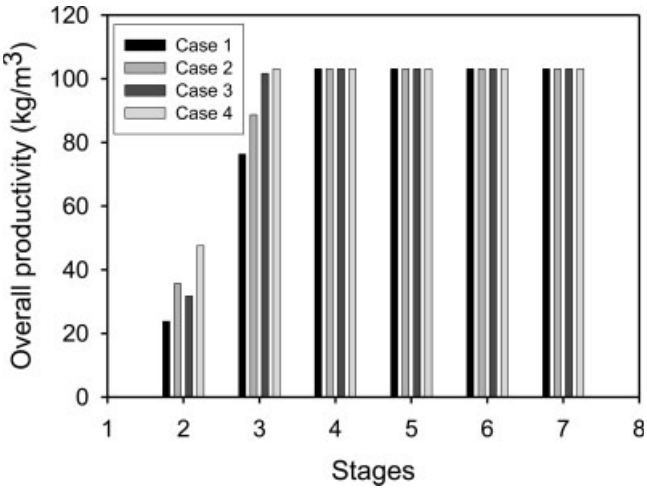

Figure 4. Overall productivity for each case under various stages.

This means that the DM would like to achieve 100% satisfaction for each objective function and inequality constraints. If the optimal decision, η_D^*, is between zero and one, then this implies that each membership function value $\eta_k(f_k)$ is also between zero and one. This situation indicates that the DM has some degree of satisfaction with the optimal solution.

The operation variables in the fuzzy goal attainment problem expressed in Eq. 31 are considered to be

$$\mathbf{z} = \left[D_1,\ s_{f_1}, \cdots s_{f_n},\ b_1 \cdots b_n,\ \alpha_2 \cdots \alpha_n,\ \beta_2 \cdots \beta_n\right] \qquad (32)$$

The HDE algorithm is applied to find an optimal design from the second stage to the seventh stage of the integrated extractive fermentation processes. For each stage, four sets of operation variables are considered as the decision variables to find the corresponding optimal design. The first set is referred to as the first case as shown in Table 2, and in which there is an equal working volume and feed flow rate for each stage, that is, $\alpha_2 = \cdots = \alpha_n = 1$ and $\beta_2 = \cdots = \beta_n = 1$. The decision variables for this set consist of the dilution rate, the feed glucose concentration and the bleed ratio for each stage, that is, $\mathbf{z} = [D_1, s_{f_1} \cdots s_{f_n}, b_1 \cdots b_n]$. The second set is referred to as the second case and an equal feed flow rate for each stage is considered. The decision variables for this set are therefore defined as $\mathbf{z} = [D_1,\ s_{f_1} \cdots s_{f_n},\ b_1 \cdots b_n,\ \alpha_2 \cdots \alpha_n]$. In the third case, the decision variables are taken to be an equal working volume for each stage, that is, $\mathbf{z} = [D_1, s_{f_1} \cdots s_{f_n}, b_1 \cdots b_n, \beta_2 \cdots \beta_n]$. The fourth case is considered for all operation variables as expressed in Eq. 32. Table 2 shows the best results for each case.

For the two-stage integrated process, we are unable to find an acceptable design for each case. The overall productivity and conversion are less than the assigned preference lower bound and the residual glucose at the final stage is greater than and equal to the upper bound. As a result, the grade of the corresponding membership function is zero. This indicates that this result is completely unacceptable to the DM. The overall productivity and residual glucose are still unacceptable for the first case in the three-stage integrated process. For this case, the conversion and total amount of supplied glucose are achieved with roughly 93% satisfaction. However, the overall grade of the membership function is zero. The second, third, and fourth cases in the three-stage integrated process achieve about 21, 48, and 51% satisfaction, respectively. When we consider a four-stage process, the best solutions for the four cases are nearly identical and attain 51% satisfaction. Similarly, the overall productivity, conversion, and total amount of supplied glucose for five-stage to seven-stage integrated processes are all nearly identical. Table 2 shows the best solutions for the seven-stage process. The optimal dilution rate for each case is different as shown in this table. Figure 4 shows the overall productivity for each case from the two-stage to seven-stage processes. The first, second, and third cases requires four stages to yield the

Table 3. Best Results for the Integrated Processes with Various Interval Boundaries for Objective Functions and Constraints

Run	$[f_1^L, f_1^U]$ (kg / m^3 h)	$[s_{f_1}^L, s_{f_1}^U]$ (kg / m^3 h)	No. of Stages	f_1^* (kg / m^3 h)	f_2^*	f_3^* (kg / m^3)	f_4^* (kg / m^3 h)	η_1^*	η_2^*	η_3^*	η_4^*	D_1 (1 / h)
1	[80, 140]	[80, 140]	2	47.5801	0.9266	0.5000	58.2193	0.0	0.0	0.0	1.0	6.0000
			3	103.0728	0.9994	0.3461	116.9272	0.5050	0.9934	0.5053	0.5050	5.9898
			4	103.0837	0.9996	0.1778	116.9163	0.5052	0.9957	0.8750	0.5052	5.9984
			5	103.1056	1.0000	0.3427	116.8944	0.5062	0.9998	0.5142	0.5056	5.9983
			6	103.1035	1.0000	0.2192	116.8965	0.5056	0.9997	0.7979	0.5056	4.8855
			7	103.1074	1.0000	0.2164	116.8925	0.5057	0.9999	0.8035	0.5057	1.6836
2	[100, 140]	[80, 140]	2	47.5800	0.8847	0.5000	60.9749	0.0	0.0	0.0	1.0	6.0000
			3	107.7694	0.9948	0.4223	122.8266	0.2793	0.9358	0.2793	0.3937	6.0000
			4	110.0856	0.9995	0.2520	124.8716	0.3526	0.9945	0.7311	0.3526	5.9774
			5	110.1095	1.0000	0.2289	124.8357	0.3533	0.9999	0.7788	0.3533	5.8595
			6	110.1083	1.0000	0.3100	124.8374	0.3533	0.9999	0.5981	0.3533	5.7453
			7	110.1102	1.0000	0.2216	124.8347	0.3533	0.9999	0.7936	0.3533	1.9923
3	[80, 140]	[100, 140]	2	47.5801	0.9266	0.5000	58.2193	0.0	0.0	0.0	1.0	6.0000
			3	107.0471	0.9951	0.3197	121.9686	0.5741	0.9396	0.5741	0.5741	6.0000
			4	107.3538	0.9996	0.2002	121.7641	0.2792	0.9953	0.8343	0.5792	5.9952
			5	107.3810	1.0000	0.0928	121.7460	0.2796	0.9999	1.0	0.5796	5.8231
			6	107.3852	1.0000	0.1474	121.7432	0.5797	0.9999	0.9268	0.5797	6.0000
			7	107.3853	1.0000	0.1635	121.7432	0.5797	0.9999	0.8999	0.5797	1.5797
4	[100, 140]	[100, 140]	2	47.5800	0.8847	0.5000	60.9749	0.0	0.0	0.0	1.0	6.0000
			3	107.7694	0.9948	0.4223	122.8266	0.2793	0.9358	0.2793	0.5522	6.0000
			4	112.4507	0.9996	0.1044	127.5493	0.4232	0.9949	0.9935	0.4232	6.0000
			5	112.4785	1.0000	0.1873	127.5214	0.4240	0.9999	0.8580	0.4240	5.8123
			6	112.4782	1.0000	0.1989	127.5218	0.4240	0.9998	0.8368	0.4239	5.7200
			7	112.4794	1.0000	0.1231	127.5206	0.4240	0.9999	0.9654	0.4240	1.6945

maximum overall productivity and nearly complete overall conversion. However, if the ratios of working volume and feed flow rate for each stage are considered as the decision variables in the optimization problem, three stages are sufficient to achieve an identical overall productivity and conversion.

Table 3 shows the best solutions to the fourth case of the fuzzy goal attainment problem with various interval boundaries for the overall productivity and supplied glucose. The first run is summarized in Table 2 as discussed above. In the second run, the DM changes the overall productivity preference such that the productivity lower bound is increased to 100 kg m^{-3} h^{-1}. The other bounds are identical to those of the first run. The second row in Table 3 shows the best results from two-stage to seven-stage processes. For the two-stage integrated process, we are still unable to find an acceptable design. The overall productivity and conversion are below the lower bounds. The residual glucose is greater than the upper bound. The three-stage problem requires 27.93% satisfaction for productivity, 93.58% for conversion, 27.93% for residual glucose, and 39.37% for supplied glucose. This run requires four stages to achieve an identical overall productivity and conversion. Similarly, the third run increases the supplied glucose lower bound to 100 kg m^{-3} h^{-1}. The other bounds are identical to those of the first run. Following a similar procedure, the three-stage problem is to obtain 57.41% satisfaction for productivity, residual glucose, and supplied glucose, and 93.96% for conversion. This run also requires four stages to achieve an identical overall productivity and conversion. The last run is to increase both overall productivity and the supplied glucose lower bound to 100 kg m^{-3} h^{-1}. The results are similar to the previous cases, as shown in the fourth row of Table 3.

Conclusions

Lactic acid is one of the most commonly fermented products. To increase the efficiency of production, various cell culture methods have been investigated. In this work, we considered a multistage integrated continuous process. Each stage consists of a mixing tank, a bioreactor, a cell-recycle unit and an extractor. The cell-recycle unit is applied to recycle the cell back into the bioreactor to yield a high cell density fermentation. The exactor is used to prevent product inhibition in the fermentation. The generalized mathematical model was formulated to express the integrated process. We have compared the overall productivity and conversion of the integrated process with those of two simplified processes. One of the simplified processes is to omit the extractor from the integrated process. The other simplified process is a series continuous fermentation, which is equivalent to omitting both the cell-recycle unit and the extractor from the proposed process. From the design equations, we see that the three processes have an identical overall conversion. However, the proposed process has the greatest overall productivity. The other processes have an identical overall productivity.

In the case study, the kinetic model of the lactic fermentation process discussed in Ben Youssef et al.[17] was applied to the integrated process to obtain the maximum overall productivity. The optimization problem was formulated as a flexible or fuzzy goal attainment problem to determine the optimal stages, operating conditions and design variables. Hybrid differential evolution was applied to solve the problem and obtain the optimal solution. The decision variables in the optimization problem consist of the dilution rate, feed glucose concentrations, the bleed ratio, the bioreactor volume ratio and the flow rate ratio for each stage. In this work, four cases of operation variables were considered as the decision variables to find the optimal design. The first case of the operation variables excludes the bioreactor volume ratio and the flow rate ratio in the optimization problem. The second case excludes the feed flow ratio rate for each stage. Third case excludes the bioreactor volume ratio. The fourth case considers all operation variables. The first, second and third cases required four stages to yield the maximum overall productivity and near complete overall conversion. However, if the ratios of working volume and feed flow rate for each stage were considered as the decision variables in the optimization problem, three stages were enough to achieve an identical overall productivity and conversion.

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Notation

b_n = bleed ratio at the nth stage bioreactor, dimensionless
D_1 = F_1/V_1, dilution rate at the first stage, h^{-1}
D_{E_n} = solvent dilution rate at the nth stage, h^{-1}
E_n = extraction efficiency at nth stage, dimensionless
F_n = feed flow rate at the nth stage, m^3 h^{-1}
F_{E_n} = solvent flow rate at the nth stage, m^3 h^{-1}
f_i = objective function and inequality constraint, $i = 1, \ldots, 4$
g_n = enrichment concentration at the nth stage, kg m^{-3}
g_{f_n} = feed enrichment concentration at the nth stage, kg m^{-3}
K = distribution efficiency, dimensionless
p_n = product (lactic acid) concentration at the nth stage, kg m^{-3}
p_{f_n} = feed product (lactic acid) concentration at the nth stage, kg m^{-3}
$r_{x_n}, r_{s_n}, r_{p_n}$ = rate equations for cell, substrate and product at the nth stage, kg m^{-3} h^{-1}
s_n = substrate (glucose) concentration at the nth stage, kg m^{-3}
s_{f_n} = feed substrate (glucose) concentration at the nth stage, kg m^{-3}
s_r^L, s_r^U = lower and upper bounds of the residual glucose, kg m^{-3}
s_t^L, s_t^U = lower and upper bounds for the total amount of glucose supplied into the process, kg m^{-3}
t = time, h
V_n = working volume in the nth stage, m^3
x_n = cell mass concentration at the nth stage, kg m^{-3}
$\mathbf{z}$ = operation variables in the optimization problem

Superscript

* = optimal solution

Greek letters

α_n = V_n/V_1, the volume ratio of the nth bioreactor to the first bioreactor, dimensionless
β_n = F_n/F_1, the ratio of the overall feed flow rate at the nth stage to that at the first stage, dimensionless
γ_n = $F_{p_{fn}}/F_1$, ratio of the product feed flow rate at the nth stage to the overall feed flow rate at the first stage, dimensionless
δ_n = $F_{s_{fn}}/F_1$, ratio of the glucose feed flow rate at the nth stage to the overall feed flow rate at the first stage, dimensionless
χ^i = overall conversion of substrate (glucose) in processes A, B, and C, dimensionless, $i = A, B, C$
χ_n^i = conversion of substrate (glucose) in processes A, B, and C at the nth stage, dimensionless, $i = A, B, C$

π^{i} = overall lactic acid productivity in processes A, B, and C, kg m^{-3} h^{-1}, $i = A, B, C$

π_n^i = lactic acid productivity in processes A, B, and C at the nth stage, kg m^{-3} h^{-1}, $i = A, B, C$

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