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Modeling and Simulation of Simultaneous Saccharification and Fermentation of Paper Mill Sludge to Lactic Acid

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Abstract Modeling and simulation for simultaneous saccharification and fermentation (SSF) process in bioconversion of paper mill sludge to lactic acid was carried out. The SSF process combined the enzymatic hydrolysis of paper mill sludge into glucose and the fermentation of glucose into lactic acid in one reactor. A mathematical modeling for cellulose hydrolysis was developed, based on the proposed mechanism of cellulase adsorption deactivation. Another model for simple lactic acid fermentation was also made. A whole mathematical model for SSF was developed by combining the above two models for cellulose hydrolysis and lactic acid fermentation. The characteristics of the SSF process were investigated using the mathematical model.

Key words: Cellulase, cellulose, paper mill sludge, lactic acid, modeling

Lactic acid has wide applications in food and pharmaceutical industries. In addition, the potential use as the source for polylactate polymers in manufacturing biodegradable plastics forms even a bigger market for lactic acid. About half of the world production of lactic acid is made from fermentation, while the remainder is produced synthetically [19]. Utilization of industry wastes, like paper mill sludge, as the substrate for fermentation of lactic acid makes the production process more attractive in both environmental preservation and production cost reduction [5, 11, 13].

About 10^6 tons of paper mill sludge are produced in Korea in each year. The dried materials comprising 40% of this sludge are composed of 30 to 60% of cellulose, which can be hydrolyzed enzymatically to produce glucose. The glucose produced can be fermented to produce lactic acid. Compared with the natural cellulosic materials like wood [16] and rice straws [7, 8], paper mill sludge needs no pretreatment before enzymatic hydrolysis, reducing the production cost. It has already been treated for removing lignin and hemicellulose in industry and is much more susceptible towards enzymatic hydrolysis [13].

The major hindrance for the commercial application of bioconversion of paper mill sludge to lactic acid was the high cost of enzymes. Thermal deactivation was ever considered a main reason [2, 3]. However, cellulase is not easily deactivated in the optimal enzymatic reaction condition of 50°C, pH 4.8, and moderate agitation. Cellulase needs to adsorb to cellulose molecules in order to react with the solid substrate. The cellulase-cellulose adsorption complex can be changed into an irreversible adsorption complex, which makes the cellulase fixed to a certain place of the surface of cellulose molecule, unable to reach new substrates and hence becomes "apparently" deactivated. The irreversible adsorption of cellulase to cellulose should be the main reason for the high cellulase consumption. Glucose and cellobiose inhibitory effects may be another reason for the high enzyme consumption. SSF can relieve the inhibitory effects by decreasing glucose and cellobiose concentrations and is a good choice for bioconversion of paper mill sludge to lactic acid [1].

With several enzymatic and one microbial reactions going on simultaneously, SSF is a relatively sophisticated process. Modeling and simulation are a powerful tool in mechanism investigation, feasibility evaluation, and process design and optimization. In this research, a mathematical model for the SSF process to bioconvert paper mill sludge into lactic acid was developed. It comprises simple cellulose hydrolysis based on the mechanism of cellulase adsorption deactivation and simple lactic acid fermentation. The characteristics of SSF were investigated by simulation, using the mathematical model.

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MATERIALS AND METHODS

Strains, Medium, and Enzymes

Raw cellulase powder [18] was produced by *Trichoderma reecei* Rut C-30 (ATCC 56765). β-Glucosidase used was the commercial product of Novozym 188 (Novo Nordisk, Denmark). Cellulose powder of type 101-F (Sigma, U.S.A.) was used in the cellulose hydrolysis experiment. Paper mill sludge (sludge for short) was obtained from Samwha Paper Co., Korea. *Lactobacillus rhamnosus* (ATCC 10863) producing L-lactic acid was used. A medium containing sodium acetate 1 g/l, K_2HPO_4 0.3 g/l, KH_2PO_4 0.15 g/l, $MgSO_4 \cdot 7H_2O$ 0.15 g/l, and glucose 85 g/l was used for lactic acid fermentation. In the SSF process, paper mill sludge was used instead of glucose.

Operation Methods

Cellulose hydrolysis was done in a 300-ml flask containing 100 ml of the mixture of cellulose power at the final concentration of 45 g/l with Na-citrate buffer (pH 4.8). After autoclaving at 121°C for 30 min and cooling down to room temperature, cellulase, measured by CMCase activity, and β-glucosidase were added to the final activities of 5 and 2 IU/ml, respectively. Oxymycin was added to the final concentration of 25 mg/ml, as an antibiotic for prevention of contamination. The flasks were incubated at 42°C and shaken at 150 rpm in a shaking water bath (KMC1205KW1, KMC Vision Co., Korea). Samples were taken for analysis of cellulose, cellobiose, and glucose.

Flask lactic acid fermentation was done using flasks of 100 ml, containing 50 ml of medium with 5% of inoculation. It was cultivated at 42°C, shaking at 150 rpm in a rotary shaking incubator of Model KMC-8480 SF (Vision Scientific Co., Korea). Four percent $CaCO_3$ were added in the medium for buffering pH.

Fermenter lactic acid fermentation was done using a 3-l fermenter (KFC, Korea) containing 1 l of medium with inoculation volume of 5%, cultivated at 42°C and agitated at 90 rpm. pH was maintained at 5.0 using 5 M NaOH solution.

SSF for lactic acid production from sludge was operated in fed-batch mode in a 3-l fermenter (KFC, Korea), containing 1 l of medium with initial sludge concentration of 15 g/l, inoculation volume of 5%, initial cellulase (measured by CMCase activity) and β-glucosidase activities of 5 and 2 IU/ml, respectively, cultivated at 42°C and agitated at 90 rpm. Sludge was fed five times at 8, 14, 20, 26, and 32 h, respectively, to increase 15 g/l sludge concentration in every feeding. Cellulase and β-glucosidase were fed two times at 14 and 26 h, respectively, to increase CMCase and β-glucosidase activities of 5 and 2 IU/ml, respectively, in every enzyme feeding. pH was maintained at 5.0 using 5 M NaOH solution.

Analytical Methods

Cellulose concentration in cellulose hydrolysis experiment was measured by the dry weight. After thorough shaking of the flask, three 1 ml aliquots of broth were accurately sampled, centrifuged, and the supernatant removed and dried at 110°C overnight until the weight was measured constant for three times. The cellulose concentration was the average of triple samples with the net error less than 5%.

Cellobiose concentration was analyzed by HPLC (Waters Co., U.S.A.) using the C18 column (Waters Co., U.S.A.) and Evaporate Laser Scattering detector (Alltech Co., U.S.A.). Glucose and lactic acid concentrations were analyzed using glucose oxidase-peroxidase and lactic acid oxidase-peroxidase methods, respectively, with an autoanalyzer (Biochemistry Analyzer 2700; YSI, Ohio, U.S.A.). Protein concentration was measured by the Lowry method [4].

Endo and exo-type glucanase activities were measured by CMCase and avicelase activities, respectively. CMCase, avicelase, and β-glucosidase activities were measured according to the method of the International Union of Pure and Applied Chemistry (IUPAC). One unit of the enzyme activity was defined as the amount of enzyme releasing 1 μmol of glucose per minute.

Cell concentration was determined by absorbance using spectrophotometry (Spectronic Instruments Co., U.S.A.) at 600 nm wavelength. Samples were diluted from 5 to 50 times to keep the value of absorbance below 0.7. The sample containing $CaCO_3$ were diluted with 0.5 M HCl solution in order to overcome the interference of $CaCO_3$ in absorbance in the measurement.

MODEL DEVELOPMENT

Cellulose Saccharification

Cellulase Adsorption It was confirmed that cellulase adsorbed to cellulose in both reversible and irreversible ways, while β-glucosidase was not adsorbed to cellulose (data not shown). Cellulase adsorption is described using the Langmuir equation:

$$E_{ads}=\frac{K_p \cdot E_{ads,m} \cdot E_L}{1+K_p \times E_L} \quad (1)$$

where E_L is cellulase concentration in the liquid phase, mg/l; E_{ads} is amount of cellulase adsorbed per gram of cellulose, mg/g; $E_{ads,m}$ is maximum amount of cellulase adsorbed per gram of cellulose, mg/g; and K_p is a constant.

$$E_L=E_0 - E_{ads} \cdot C \quad (2)$$

where, E_0 is the total cellulase concentration, mg/l; and C is cellulose concentration, g/l.

The adsorbed cellulase, E_{ads}, is converted to the irreversibly adsorbed cellulase-cellulose complex. The reversibly adsorbed cellulase, E_a, decreases in the following way:

$$\frac{dE_a}{dt}=-k_d \cdot E_{ads} \cdot C \quad (3)$$

where k_d is a constant, 1/h.

Enzymatic Reaction Kinetics The irreversibly adsorbed cellulase was fastened to a fixed place in the surface of the cellulose molecule, unable to reach new substrate of cellulose, and was "apparently" deactivated. Only reversibly adsorbed cellulase could react with cellulose and the reaction rate is as follows:

$$r_1=\frac{K_1 \cdot E_a \cdot C}{K_{1s} \cdot (1+B/K_{1B})+C} \quad (4)$$

where r_1 is the reaction rate of cellulose, K_1, K_{1s}, K_{1B} are constants, and B and C are the concentrations of cellobiose and cellulose.

The fact that a small amount of cellulase liquefied cellulose with little glucose produced (data not shown) showed that the substrate of exoglucanase, which is an oligosaccharide, was soluble. For soluble substrate, the adsorbed cellulase was a kind of immobilized enzyme, having much decreased activity compared with the soluble state enzyme. As a result, only the soluble state cellulase was counted on the oligosaccharide conversion with the reaction rate as follows:

$$r_2=\frac{K_2 \cdot E_L \cdot OS}{K_{2s} \cdot (1+B/K_{2B})+OS} \quad (5)$$

where r_2 is the reaction rate of oligosaccharide, OS is the concentration of oligosaccharide. K_2, K_{2s}, K_{2B} are constants.

$$r_3=\frac{V_{3m} \cdot B}{K_{3s} \cdot (1+G/K_{3G})+B} \quad (6)$$

where r_3 is the reaction rate of cellobiose, V_{3m}, K_{3s}, and K_{3G} are constants, and G is the concentration of glucose.

Mass Balances for the Reactions

$$\frac{dC}{dt}=-r_1 \quad (7)$$

$$\frac{dOS}{dt}=r_1-r_2 \quad (8)$$

$$\frac{dB}{dt}=1.056 \cdot r_2-r_3 \quad (9)$$

$$\frac{dG}{dt}=1.053 \cdot r_3 \quad (10)$$

Lactic Acid Fermentation

The specific growth rate, μ, and the specific lactic acid production rate, q_P, are described using Equations (11) and (12), respectively:

$$\mu=\frac{\mu_{max} \cdot G}{k_m+G} \cdot \frac{k_{iG}}{k_{iG}+G} \cdot \left(1-\frac{P}{P_{cri}}\right)^n \cdot \left(1-\frac{X}{X_{max}}\right) \quad (11)$$

$$q_P=\alpha \cdot \mu+\beta \quad (12)$$

The byproducts of oligosaccharides and other kinds of organic acids in addition to lactic acid were found in fermentation broth (data not shown). The specific byproduct production rate, q'_P, was found to be growth related:

$$q'_P=\gamma \cdot \mu \quad (13)$$

The specific glucose consumption rate for byproduct synthesis, q'_G, is as follows:

$$q'_G=\frac{q'_P}{Y'_{P/G}}=\frac{\gamma \cdot \mu}{Y'_{P/G}}=\frac{1}{\delta} \cdot \mu \quad (14)$$

where $\delta=Y'_{P/G}/\gamma$. The total specific glucose consumption rate, q_G, is:

$$q_G=-\frac{\mu}{Y_{X/G}}-\frac{q_P}{Y_{P/G}}-q'_G=-\left(\frac{1}{Y_{X/G}}+\frac{1}{\delta}\right) \cdot \mu-\frac{q_P}{Y_{P/G}} \quad (15)$$

In complex medium, carbon source is used for only energy production and products synthesis, not for cell synthesis, which was also confirmed using *Lactobacillus rhamnosus* [15, 20]. In this research, complex medium was utilized, therefore, the term of glucose consumption for cell synthesis in Equation (15) was omitted in the following simulations.

The mass balances for cell, lactic acid, and glucose are expressed by Equations (16)-(18):

$$\frac{dX}{dt}=\mu \cdot X \quad (16)$$

$$\frac{dP}{dt}=q_P \cdot X \quad (17)$$

$$\frac{dG}{dt}=-q_G \cdot X \quad (18)$$

Simultaneous Saccharification and Fermentation

In SSF, glucose produced in sludge hydrolysis was simultaneously consumed by the microorganisms. Combining the equations for glucose production (10) and glucose consumption (15) and (18), Equation (19) was built for modeling the dynamics of glucose concentration in SSF:

$$\frac{dG}{dt}=1.053 \cdot r_3-\frac{\mu \cdot X}{\delta}-\frac{1}{Y_{P/G}} \cdot \frac{dP}{dt} \quad (19)$$

In modeling SSF, Equations (1)-(9), (11), (12), (15)-(17), and (19) were used. Besides, the SSF was operated in fed-batch mode in experiments, so that the equations were modified for the dilution effect caused by the substrate feeding in the SSF simulation. However, the dilution effect caused by enzyme feeding was neglected.

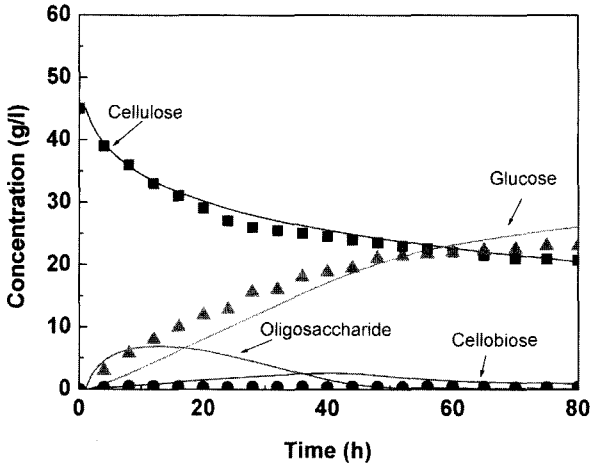

Fig. 1. Time course of cellulose hydrolysis.
The scattered symbols are experimental data, and the lines are model predictions.

RESULTS AND DISCUSSION

Cellulose Hydrolysis

Cellulose hydrolysis experiment was performed and the time courses of cellulose, cellobiose, and glucose concentrations were measured (Fig. 1). Glucose concentration reached about 24 g/l at around 50 h from the start of hydrolysis, and did not increase much from 50 to 80 h. Cellobiose concentration was maintained at low level all the time.

The model Equations (1)-(10) were used in the simulation of cellulose hydrolysis. The differential equations of the mathematical model were solved using the fourth-order Runge-Kutta method [10]. The parameter $E_{ads.m}$ was measured under various cellulase concentrations, and K_P was obtained by linear plots using the experimental data. For the determination of the values of V_{1m}, K_{1S}, K_{1B}, V_{3m}, K_{3S}, and K_{3G}, experiments on cellulose and cellobiose hydrolysis using cellulase and β-glucosidase, respectively, were done using various substrate concentrations, and the experimental data were plotted to obtain the values of the above parameters. K_1 was calculated using V_{1m} and the protein concentration of cellulase. For the determination of the values of K_{1B} and K_{3G}, experiments on cellulose and cellobiose hydrolysis were performed at various concentrations of the inhibition of cellobiose and glucose, respectively. The data of r_1 and r_3 versus the respective inhibition concentration were fitted with Equations (4) and (6), respectively, using nonlinear fitting to obtain the constant values. The values of K_2, K_{2S}, K_{2B}, and k_d were obtained using parameter optimization using the experimental data of the time course of cellulose hydrolysis. Parameter optimization was done automatically by computer through search of the parameter values that minimized the sum of the squared errors between model prediction and experimental data using genetic algorithm (GA) [6]. All the parameter values were then refined using GA from the experimental data of SSF for lactic acid production from sludge, as described later in this paper. The final values of model parameters are shown in Table 1. All the calculations were carried out on an IBM compatible computer, using self programmed Visual Basic (Microsoft Co., U.S.A.) programs.

Table 1. Parameter values for mathematical model of cellulose hydrolysis.

Parameter	Value	Unit
$E_{ads.m}$	98.2900	mg/g
K_p	0.1855	l/mg
k_d	1.4698	1/h
K_1	4.1711	g/mg/h
K_2	0.4156	g/mg/h
V_{3m}	2.0000	g/l/h
K_{1S}	9.4647	g/l
K_{2S}	0.1161	g/l
K_{3S}	0.3078	g/l
K_{1B}	5.9097	g/l
K_{2B}	6.7788	g/l
K_{3G}	0.9455	g/l

The mathematical model fitted satisfactorily with the experimental data (Fig. 1), supporting the hypothesis of the mechanism of cellullase adsorption deactivation. This model can be used to predict the effects of both reversible and irreversible adsorption of cellulase and the effects of the portions of endo and exo-type glucanase as well as β-glucosidase on cellulose hydrolysis, to provide useful information for process optimization.

Lactic Acid Fermentation

Lactic acid fermentation at various initial glucose concentrations, ranging from 10 to 50 g/l, was done using flask culture, and the μ_{max}, k_m, and k_{iG} parameters were calculated. The result showed that glucose inhibition was moderate. Lactic acid fermentation at various initial lactic acid concentrations from 0 to 90 g/l was done using flask, culture and the specific growth rates were calculated and plotted (Fig. 2). It shows that lactic acid inhibition was severe: Initial lactic acid concentration of 30 g/l decreased the specific growth rate by 21 percent from 0.48 to 0.38 1/h and was compared with the case with no initial lactic acid. Almost no cell growth was found in the case of initial lactic acid concentration of 70 g/l, which was defined as the critical lactic acid concentration, P_{cri}. The specific growth rate was fitted with the experimental data, and the parameter value of n of Equation (11) was determined to be 0.7. The parameter values of α, β, δ, and $Y_{X/S}$ were calculated using the data of the above two experiments at various initial glucose and lactic acid

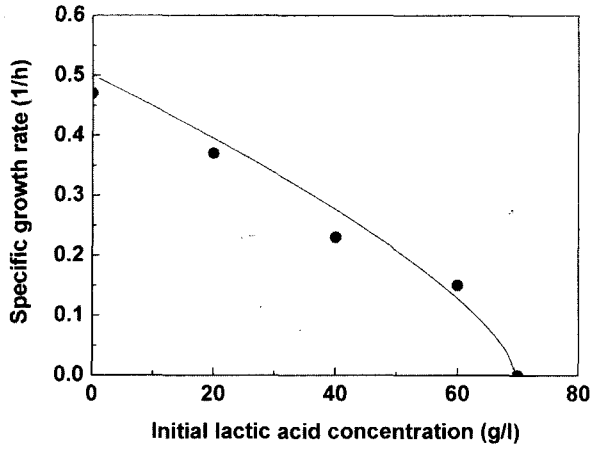

Fig. 2. Effects of initial lactic acid concentration on the specific growth rate of *Lactobacillus*.
The scattered symbols are experimental data, and the lines are model predictions.

concentrations. The value of δ varied very much with the cultivation conditions. It was about 0.7 for cultivation using flasks and was about 0.3 for cultivation using fermenter. The value of $Y_{P/S}$ was obtained by theoretical calculation, based on the fact that two lactic acid molecules are produced from one glucose molecule ($Y_{P/S}=(2\times M_{Lac})/M_{Gluc}=1$g/g). The parameter value of X_{max} was directly measured.

Lactic acid fermentation was done using fermenter, and the experimental data were used in refinement of the parameter values of the mathematical model for lactic acid fermentation using GA (Fig. 3). Then, the parameter values were further refined using the experimental data of SSF for lactic acid production from sludge. However, no refinement was made for the theoretical parameter value of $Y_{P/S}$. The parameter values for lactic acid fermentation

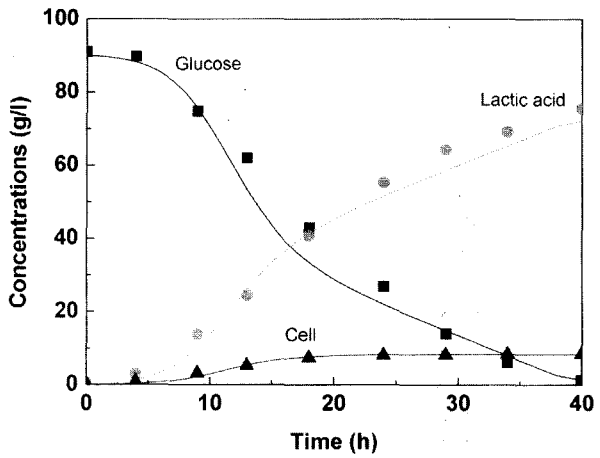

Fig. 3. Time course of lactic acid fermentation using glucose.
The scattered symbols are experimental data, and the lines are model predictions.

Table 2. Parameter values for mathematical model of lactic acid fermentation.

Parameter	Value	Unit
μ_{max}	0.576	1/h
k_m	0.3	g/l
P_{crt}	70	g/l
k_{iG}	0.2	g/l
n	0.7	-
$Y_{P/S}$	1.0	g/g
α	5	-
β	0.09	-
δ	0.286	g/g

mathematical model are shown in Table 2. Simulation for fermenter lactic acid fermentation was made, and the simulation results were compared with the experimental data, as shown in Fig. 3. The simulation fitted satisfactorily with the experimental data.

In modeling cell growth, the term of $(1-X/X_{max})$ from logistic equation was used for the stationary growth phase, even if there was other kind of equations [12]. The Luedeking-Piret model [14] was used in modeling lactic acid production. This model was simple and practical.

Simultaneous Saccharification and Fermentation

Lactic acid production from sludge using SSF was done in fed-batch mode using fermenter. During the SSF process, the enzymes were fed 2 times at 8 and 14 h, respectively, and sludge was fed 5 times at 8, 14, 20, 26, and 32 h, respectively. The results are shown in Fig. 4. The final lactic acid concentration reached the highest value of 63 g/l at about 40 h from the start of cultivation. Glucose concentration had a small increase at 4 h, reaching the

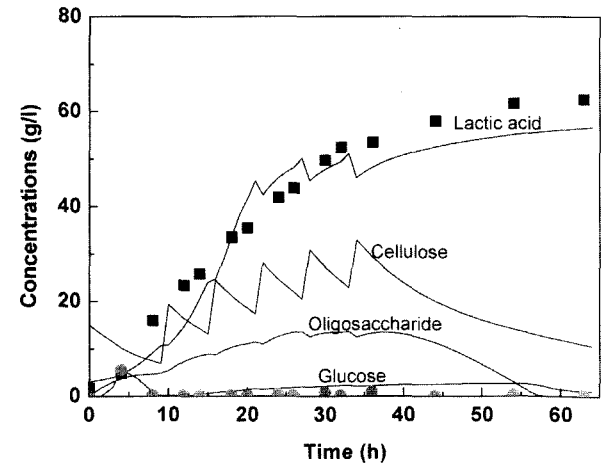

Fig. 4. Time course of fed-batch SSF in production of lactic acid using sludge.
The scattered symbols are experimental data, and the lines are model predictions.

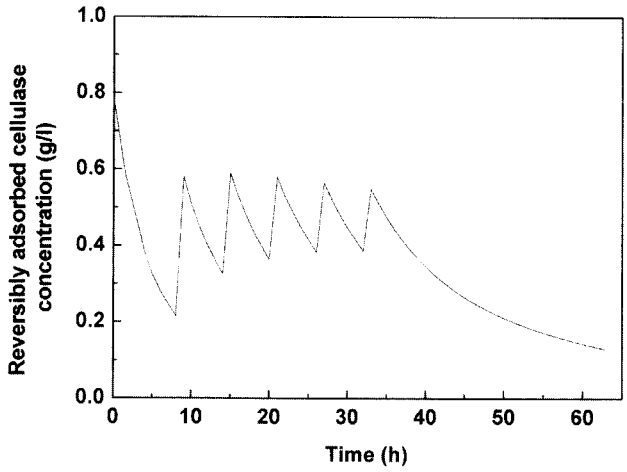

Fig. 5. Time course of predicted reversibly adsorbed cellulase during the fed-batch SSF.

highest value of 5 g/l, but soon decreased with the cell growth and remained at low level during the later period of the cultivation. Simulation of the SSF process was made using the parameter values listed in Tables 1 and 2, and the results are shown in Figs. 4–6. The model simulation fitted satisfactorily with the experimental data (Fig. 4).

The simulation results showed that the reversibly adsorbed cellulase decreased quickly due to conversion to irreversibly adsorbed form (Fig. 5). The curve of the reversibly adsorbed cellulase concentration of a saw-tooth shape resulted from the feedings of sludge as well as enzyme. Simulation of the rates of cellulose hydrolysis is shown in Fig. 6. The time course of r_1 was also of saw-tooth shape, almost parallel with the curve of the concentration of reversibly adsorbed cellulase as expected. r_2 and r_3 were catalyzed by soluble state enzymes, as analyzed above, therefore, the curves of r_2 and r_3 were quite different from

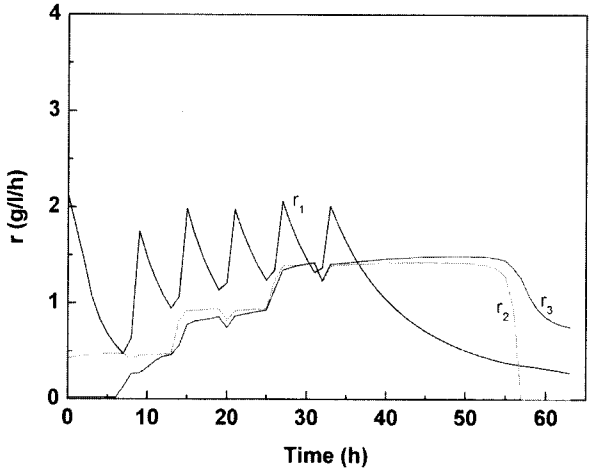

Fig. 6. Time course of predicted enzymatic reaction rates of r_1, r_2, and r_3 during the fed-batch SSF.

r_1. The simulation showed that the reaction rates, especially r_1, were greatly decreased due to irreversible adsorption of cellulase. It was expected that increase of the reversible and decrease of the irreversible adsorption of cellulase by modification of the binding domain through recombinant gene technology could greatly increase the cellulose hydrolysis efficiency.

Conventionally, it is regarded that the higher the affinity of cellulase to cellulose the better for the reaction. As discussed earlier, as cellulase has no mechanism to move along the surface of cellulose, it needs to adsorb, desorb, and re-adsorb to or from the cellulose surface, so that it can reach new substrates and react continuously. Irreversibly adsorbed cellulase fails to reach new substrates and is apparently deactivated, even though the enzyme reaction domain still has activity. It was confirmed experimentally that about 8.8% of the total adsorbed cellulase were changed into irreversibly adsorbed form (data not shown). Using atomic force microscopy, the above hypothesis was supported by the experimental results that many holes were left on the surface of cellulose molecules after co-cultivation with the reaction domain deactivated cellulase [9]. It showed that the cellulase binding domain penetrated deeply into the cellulose molecule, which might lead to the formation of the tightly combined cellulase-cellulose complex and irreversible cellulase adsorption.

Lactobacillus also has the ability to ferment cellobiose to produce lactic acid. Experiments on lactic acid fermentation, using cellobiose and the mixture of cellobiose and glucose, showed that cellobiose utilization was repressed by glucose (data not shown). Furthermore, β-glucosidase activity was high, and the result of the cellulose hydrolysis experiment showed that cellobiose concentration was at a low level (Fig. 1). Therefore, cellobiose fermentation to lactic acid by *Lactobacillus* was omitted in the SSF mathematical model.

In conclusion, SSF is a good choice for bioconversion of cellulosic materials to lactic acid. The conditions for cellulose hydrolysis and lactic acid fermentation are similar, which favors the two reactions to be carried out simultaneously in the same reactor and greatly simplifies the production process. Besides, SSF can relieve inhibition effects of glucose and cellobiose [1]. Utilization of paper mill sludge instead of natural cellulosic materials saves not only the pretreatment cost, but also relieves the environmental pollution problem of paper industries. Modeling and simulation are useful in investigation of characteristics, analysis, as well as optimization of the SSF process.

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NOMENCLATURE

B : cellobiose concentration (g/l)
C : cellulose concentration (g/l)
E_0 : total cellulase concentration (mg/l)
E_a : reversibly adsorbed cellulase concentration (mg/l)
E_{ads} : amount of cellulase adsorbed per gram of cellulose (mg/g)
$E_{ads,m}$: maximum amount of cellulase adsorbed per gram of cellulose (mg/g)
E_L : cellulase concentration in liquid phase (mg/l)
G : glucose concentration (g/l)
K_1 : constant (g/mg/h)
K_{1B} : cellobiose inhibition constant for r_1 (g/l)
K_{1s} : cellulose saturation constant (g/l)
K_2 : constant (g/mg/h)
K_{2B} : cellobiose inhibitory constant for r_2 (g/l)
K_{2s} : oligosaccharide saturation constant (g/l)
K_{3G} : glucose inhibitory constant for r_3 (g/l)
K_{3s} : glucose saturation constant (g/l)
k_d : conversion constant of adsorbed cellulase from reversible to irreversible form (1/h)
k_{iG} : glucose inhibitory constant for *Lactobacillus* cell growth (g/l)
k_m : glucose saturation constant for *Lactobacillus* cell growth (g/l)
K_p : constant of Langumir equation (l/mg)
M_{Lac} : molecular weight of lactic acid (g/mol)
M_{Gluc} : molecular weight of glucose (g/mol)
n : constant (-)
OS : oligosaccharide concentration (g/l)
P : lactic acid concentration (g/l)
P_{cri} : critical lactic acid concentration for inhibition of glucose consumption (g/l)
q_G : total specific glucose consumption rate (g/g/h)
q'_G : specific glucose consumption rate for byproduct synthesis (g/g/h)
q_P : specific lactic acid production rate (g/g/h)
q_P : specific byproduct production rate (g/g/h)
r_1 : reaction rate of cellulose (g/l/h)
r_2 : reaction rate of oligosaccharide (g/l/h)
r_3 : reaction rate of cellobiose (g/l/h)
X : cell concentration (g/l)
X_{max} : maximum cell concentration (g/l)
$Y_{P/G}$: lactic acid yield from glucose (g/g)
$Y_{X/G}$: cell yield from glucose (g/g)

Greek Symbols

α : constant (g/g)
β : constant (g/g/h)
δ : constant (g/g)
γ : constant (g/g)
μ : specific growth rate (1/h)
μ_{max} : maximum specific growth rate (1/h)

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