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Full Length Research Paper

Stimulation of fermention with uncooked rice by kinetics models and genetic algorithm

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Genetic algorithm was used to stimulate the fermentation process of ethanol fermentation with uncooked rice, genetic algorithm was designed to simulate biologic evolution process of natural system, genetic algorithm was a random intelligent exploitation within a defined search space. This algorithm was used to exploit parameters of two equations which were deducted by Verhulst, Aiba and Moser performed to depict exponential growth phase of yeast to stimulate fermentation model of exponential growth phase. The yeast growing in the proper growth phase was inoculated to medium with raw material, Dry weight, ethanol concentration and substrate consume were measured in the process of fermenting with uncooked materials. Equations deduced by Aiba and Moser were used to set up the fitness function, and the value of this function infinitely approached zero. With measured data, parameters of fitness function were deduced by Aiba and Moser. Fitted values were gained through solving the fitted equations by Runge-Kutta method and the errors between measured values and fitted values were also evaluated by drawing the fitted curve and the actual curve. Solving those fitted equations, parameters of fermentation process could be deduced.

Keywords: algorithm, dry weight, ethanol concentration, substrate concentration

INTRODUCTION

Fermentation processes product many substances in chemical and food industries, as we know the traditional fermentation is a sophisticate process, which has high cost and energy consumption. (Qi and Wang, 1999) The alcohol fermentation with uncooked material meets several needs, which include low cost, low level contaminant and rapid fermentation rate.

Genetic algorithm simulates inheritance and evolutionary process to become a self-adapting global optimization probability search algorithm (Holland, 1975), genetic algorithm is a stochastic global search method that imitates principles of natural biological evolution (Chen et al., 2004). Genetic algorithm can effectively search in complex parameter space; it can solve the problems which have too many parameters to be calculated by the conventional method. In optimizing multi-parameter nonlinear problems, this method creates initial points then does the parallel search within the solution space; consequently gets the global optimum by a biggish probability (Rivera et al., 2006).

In this experiment, genetic algorithm was preformed to optimize fermentation parameters and stimulate fermentation process. Depicting exponential growth phase of yeast cultured in wort with Verhulst equation, we obtained the fitted Verhulst equation that could stimulate yeast fermentation model of exponential growth phase by genetic algorithm. Models deduced by Aiba and Moser were performed to simulate brewing process of uncooked materials. Dry weight, ethanol concentration and substrate consume were measured from brewing with uncooked materials, then parameters of kinetics models were calculated.

Fitted values were gained through solving the fitted equations by Runge-Kutta method and the error between measure values and fitted values was also evaluated by drawing the fitted curve and the actual curve. Using those fitted equations, parameters of fermentation process could be deduced, so the model of fermenting process was known. By substitution of the initial substrate concentration and biomass into the fermenting model which had parameters deduced by fitness equation, parameters of any time of the fermentation process could be predicted by this model, such as the amount of product, terminal time of fermentation.

Experiment steps

Raw materials

Rice and bran were used in our experiment; rice contains 74.6% starch and 13.7% water; bran contains 8% water. Other additives are liquefaction enzyme (Botang, Chian) and glucoamylase (Botang , china).

Alcohol fermentation with wort

Inoculated yeast (AnQi China) into seven 250mL erlenmeyer flasks respectively, added 100 ML wort medium (130.1g/L, Siya company China) into each erlenmeyer flask. Cultivated at 30℃, separated the yeast every 6 hours by filtrating to abstain the yeast dry weight. The dry weight was total biomass, generally the dry weight is 20%-25% of green weight.

Alcohol fermentation of raw material

100g rice power, 0.11g yeast (dry yeast, AnQi China), 320mL water, 0.2g liquefaction enzyme (Liquefaction enzyme activity is 10000u/g) and 0.16g glucoamylase (glucoamylase activity is 53000u/g), were added into 500mL erlenmeyer flasks, then cultured at 32□ for 12 days. Every 12 hours, measured the concentration of biomass (g/L), the concentration of fermentable glucide (g/L) and the alcoholicity (ml/L) at 20□ of fermenting mash.

RESULTS AND ANALYSIS

Estimating the yeast fermentation kinetic model parameter cultured with wort

Experiment data

Result was listed in table 1. Repeated afore-mentioned experiment steps of 2.2, and measured the biomass at every 6 hours.

Fermentation model of culturing with wort in exponential growth phase

The process of yeast exponential growth was fit for the Verhulst equation, which depicts the growth of yeast population (Zhuge and Wang, 1994).

$$\frac{dy}{dt} = r*y*(1-y/k)\ldots\ldots\ldots y_0 = y(0) \tag{3.1}$$

After solving the differential equation, equation (3.2) was obtained as follow:

$$y(t) = k/[1+\exp(a-r*t)] \tag{3.2}$$

Formula (3.2) could be used for the fermentation kinetic model of yeast in the fermenter, and k、a、r were pending parameters.

Program of genetic algorithm (Wang, 1994)

1) Designing and coding the adaptive value f of the genetic algorithm as follow:

$$f = 1/(S+1);\quad \mathrm{S} = \sum_{i=1}^{n}(y_i - \hat{y}_i)^2 \tag{3.3}$$

In this equation S was sum of square deviation, y_i was the actual value of cell concentration obtained at the ith

Table1. Biomass in fermentation process. Yeast comes into stationary phase after 24 hours'

cultivation time (h)	biomass (g/L)
0	0
6	0.23
12	0.72
18	2.01
24	2.38
30	2.54
36	2.542

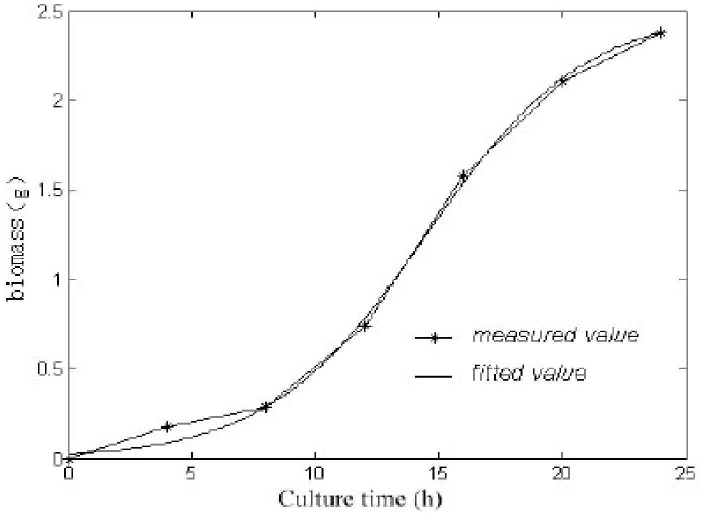

Figure 1. The curves of experimental values and model predictive values (fitted values). Fitted values were derived from the algorithm. Curve drawn with measured values (*) were approximate the curve, evaluating from this figure. Fitted curve was good to simulate the measured one.

measure, $\hat{y}_i$ was predictive value of cell concentration at the ith measure.

2) In genetic algorithm, the population scale N_p= 400,genetic generations genm= 200. Domain of k 、a 、r was [0,5].

3) Created the initial population gen =100.

4) Used the adaptive function (3.3) to computing adaptive value for every individual.

5) Carried out the program by matlab 7.

6) If the computing result met termination condition, get the optimal solution. If the computing result did not meet termination condition, the steps should return to step 4) and supposed gen =gen + 1.

The parameter of fermentation model

Assumed initial populations were 100, mutation probability was 0.08, crossover probability was 0.9, then $k = 2.5003$; $a = 4.5411$; $r = 0.3136$; $f = 0.9876$ were derived from the algorithm. Unknown parameters of equation (3.2) were substituted the by calculated values, solving the equation by Runge-Kutta, we got the fitted values and measured values. Curves of those two kinds of values were drawn in Figure 1. Results show that fitted curve was good to simulate the measured one. Result of evaluating the algorithm was shown in Figure 2, which illustrated that when the result closed to 150 generations value of optimal adaptation approached average of fitness value and the algorithm constringed rapidly, so this algorithm was effective.

Estimating the parameters from kinetic model of fermenting with raw material

Results of the yeast biomass, cell growth rate, alcohol volume and the substrate concentration in brewing with uncooked material.

We can replace the starch content by the reducing sugar (in the fermentation process, starch is decomposed to

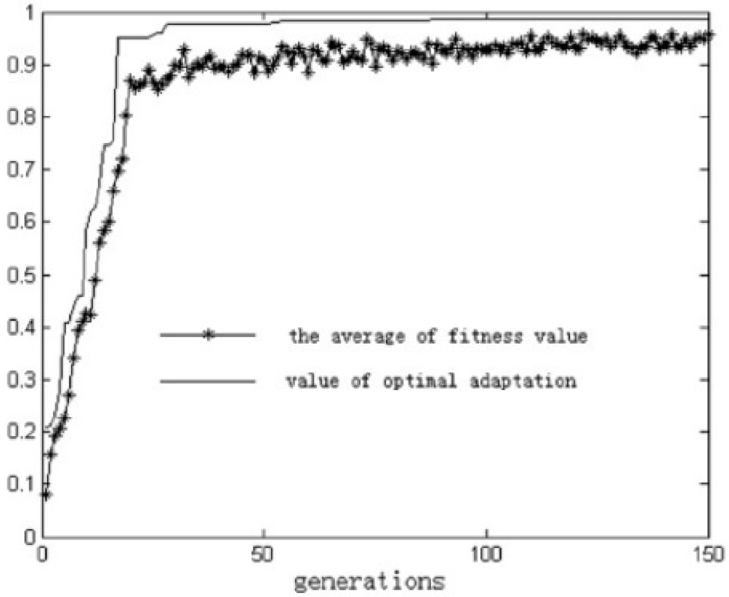

Figure 2. Evaluation chart of algorithm. when the genetic algorithm progressed closely to 150 generations, value of optimal adaptation approached average of fitness value (*) and the algorithm constringed rapidly, so this algorithm was effective.

Table 2 Rults of brewing with raw material

time (h)	biomassCi(g/L)	alcohol concentration(ml/L)	substrate concentration(g/L)
0	0.10	0	191.8
12	0.18	1.4	190.2
24	0.23	2.8	185
36	0.36	4	181.9
48	0.63	6.79	165
60	0.86	12.7	150.7
72	1.5	14.6	127.8
84	1.81	24.9	122.6
96	1.85	29.8	107.8
108	2.02	39.2	105.9
120	2.04	58.29	101.7
132	2.2	65.3	83.9
144	2.27	73.7	53.3
156	2.31	78.1	41.1
168	2.33	83.6	3.57
180	2.24	86.82	3.12

reducing sugar), due to the concentration of reducing sugar in fermenting process was higher than the concentration yeast need and the lowest concentration which can restrain yeast growth. Yeast biomass, cell growth rate, alcohol volume and the substrate concentration were shown in Table 2.

Model of yeast growth, substrate consume and ethanol production

1) Growth rate of yeast was calculated by the equation as follows:

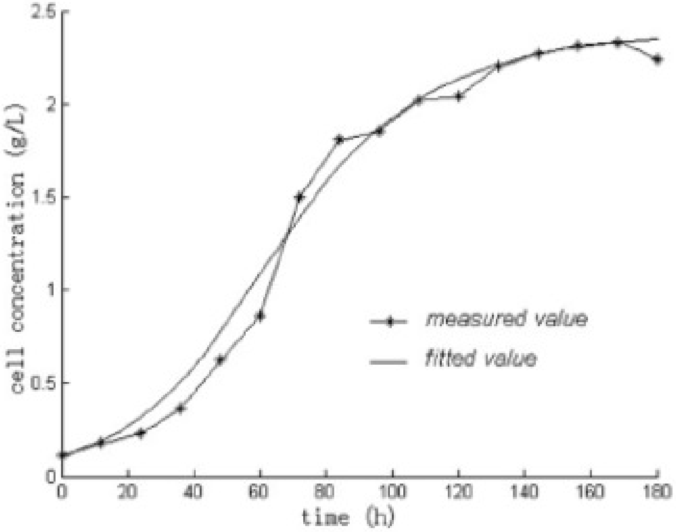

Figure 3. Concentration of biomass in fermenting with raw material. Biomass of any time in the fermentation process was deducted through solving fermentation kinetics models by Runge-Kutte method. Without an optimal method to check the accuracy of genetic algorithm results, it's a relatively good method to draw the curve contrasting with the fitted curve. Curve of the fitted values approached measured curve.

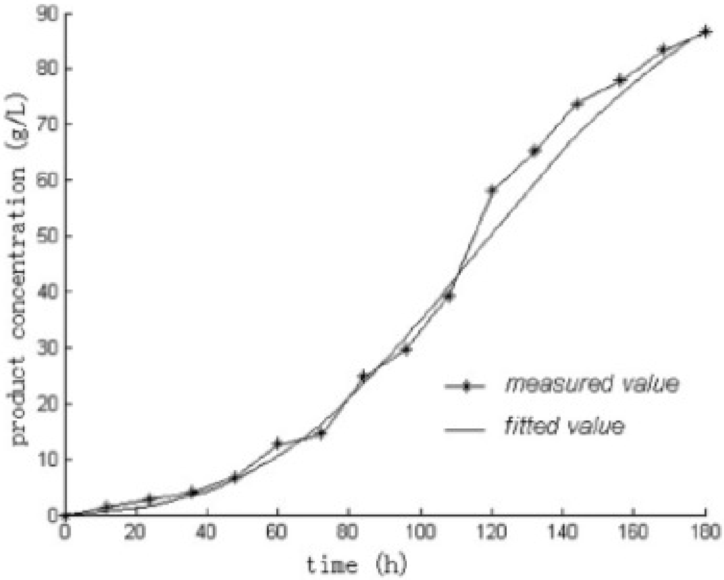

Figure 4. Ethanol Concentration in fermenting with raw material. Production concentration of any time in the fermentation process was deducted through solving fermentation kinetics models by Runge-Kutte method. Drawing the curves of measured values (*) and the fitted one, we found the fitted one could simulated the fermenting process.

$$\mu_i = (X_{i+1} - X_i) / \Delta t \tag{3.4}$$

X_i: The dry weight of biomass at the i th measure. Following function was used to calculated specific growth rate of yeast:

$$v_i = (P_{i+1} - P_i) / \Delta t \tag{3.5}$$

P_i: The alcohol concentration of the i th measure.

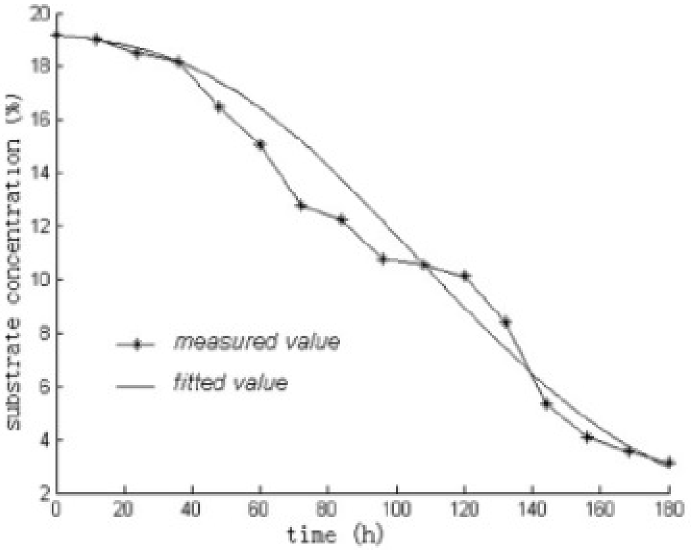

Figure 5. Glucose concentration in fermenting with raw material.

Specific consumption rate of substrate was expressed by (3.6):

$$m_i = (S_{i+1} - S_i) / \Delta t \qquad (3.6)$$

The amount of substrate consumption of the i th measure.

2) Model of alcoholic fermentation

Since alcohol fermentation is complex processes, besides the substrate consumption and the alcohol production, this process also has breath, cell growth and metabolism etc. At present, fermentation kinetics is divided into three parts: 1. model of cell growth dynamics, 2. model of substrate consumption kinetic, 3. kinetic model of production dynamics.

3) Model of yeast growth dynamics

The definition of yeast growth (3.7) and alcohol production models (3.8) are:

$$\frac{dX}{dt} = \mu X \qquad (3.7)$$

$$\frac{dP}{dt} = vX \qquad (3.8)$$

Ethanol production by yeast fermentation accords with product inhibition model. In the product inhibition models, the most representative one is the model designed by Aiba and Moser. (Qi and Wang, 1999), (Shule and Kargi, 2002), (Shagen, 1998).

$$\mu = \frac{\mu_{\max} * S * K_P}{\left(K_S + S + S^2/K_{SI}\right) * \left(K_P + P + P^2/K_{PI}\right)} \qquad (3.9)$$

$$v = \frac{v_0 * S * K'_P}{\left(K'_S + S + S^2/K'_{SI}\right) * \left(K'_P + P + P^2/K'_{PI}\right)} \qquad (3.10)$$

$$\frac{dS}{dt} = -\left(\frac{\mu}{K_{X/S}} + \frac{v}{K_{P/S}}\right) * X \qquad (3.11)$$

Hereinbefore the symbols of μ 、v 、S 、P 、X were respectively biomass specific growth rate, specific generation rate of product, substrate concentration, product concentration, biomass concentration.

Parameters of fermentation dynamics model in fermenting with raw material

Equations (3.12) and (3.13) deducted by the above equations (3.7), (3.8), (3.9), (3.10) and (3.11).

$$\frac{dX}{dt} = \frac{\mu_{\max} * S * K_P * X}{\left(K_S + S + S^2/K_{SI}\right) * \left(K_P + P + P^2/K_{PI}\right)} \qquad (3.12)$$

$$\frac{dP}{dt} = \frac{v_0 * S * K'_P * X}{\left(K'_S + S + S^2/K'_{SI}\right) * \left(K'_P + P + P^2/K'_{PI}\right)} \qquad (3.13)$$

$$\frac{dS}{dt} = -\left(\frac{\mu}{K_{X/S}} + \frac{v}{K_{P/S}}\right) * X \qquad (3.14)$$

Fitness function was defined as follow:

$$J = \sum ((\frac{dX}{dt} - Y_X)^2 + (\frac{dP}{dt} - Y_P)^2 + (\frac{dS}{dt} - Y_S)^2) \qquad (3.15)$$

Y_X 、Y_P 、Y_S were the computed value of the dexter equation(3.8), (3.9) and (3.10) respectively. The genetic algorithm performed by matalab was used to obtain the maximum value of objective function. But in the equation (3.15) $J \to 0$, so we established the objective function f:

$$f = 1/(1+J) \tag{3.16}$$

$\mu_{\max}$ was the maximum specific growth rate of biomass; K_p was product saturation constant; K_s was substrate saturation constant of biomass; K_{SI} was substrate inhibition constant of biomass; K_{PI} was product inhibition constant of biomass; v_0 was the maximum specific production rate of ethanol; K'_P was product saturation constant; K'_S was the substrate saturation constant of ethanol; K'_{SI} was substrate inhibition constant of ethanol; K'_{PI} was the product inhibition constant of ethanol, $K_{X/S}$ is coefficient biomass yield vs. substance consumption; $K_{P/S}$ is coefficient of ethanol yield vs. substance consumption. Equation (3.16) has 12 unknown parameters with their bounds defined from 0 to 100. For each parameter, mutation probability was 0.1, and crossover probability was 0.9.

If calculating results of any parameter was between 99and 100, the bounds of value range.we should be adjusted. Providing that K_s =98, we substituted the area by [50 150]. Finally, when the values of 12 parameters were not approximate with their upper or lower bound, we can trust those results. Parameters were calculated as follow:

$\mu_{\max} = 0.6851$; $K_P = 18.0717$; $K_S = 145.9279$;
$K_{SI} = 3.0668$; $K_{PI} = 24.569$
$v_0 = 3.9293$; $K'_P = 6371.7$; $K'_S = 47.1035$;
$K'_{SI} = 2.5898$; $K'_{PI} = 3.8072$
$K_{X/S} = 0.9374$; $K_{P/S} = 6.2597$

Checking the results of genetic algorithm

Substituted the unknown parameters of (3.12)、(3.13)、(3.14) by caculated values and initial values: $X_0 = 0.11$; $S_0 = 19.18$; $P_0 = 0$, and solve them by Runge-Kutta. The diagram with the values of actual measurement and the values solved from (3.12)、(3.13)、(3.14) was made. Variation of the cell concentration, ethanol concentration, and substrate concentration was shown in Figure 3, Figure 4 and Figure 5 respectively.

CONCLUSION

In the Figure 3, 4 and 5, curves of the three measured values approached their own fitted curve, so the results of (3.12), (3.13) and (3.14) were appropriate. Without an optimal method to check the accuracy of genetic algorithm results, it's a relatively good method to check the parameters by contrasting the measured curves and curves of fitted values. After substituting the values of parameters, the kinetics model of fermentation with uncooked rice could be abtained. Those fermentation kinetics models are suitable for predicting the parameters of fermentation process. Product concentration, substrate concentration and biomass at any time of the fermentation process were deducted through solving fermentation kinetics models by Runge-Kutte method when the initial values of substrate concentration and biomass were gained.

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