Optimal design of automatic control system for the fed-batch production of 1,3-propanediol with pH logic control

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In this work, the fed-batch fermentation of glycerol by \textit{Klebsiella pneumoniae} with pH logic control is discussed. We propose a hybrid system constrained optimal control problem with glycerol feeding instants as control variables to maximize the concentration of the objective product at the terminal time. Correspondingly, we explore an automatic control system to implement the optimal control strategy, in which the designs of control diagram, PLC control and supervisory computer control system are involved. This work achieves a goal of applying numerical optimal control analysis to the design of control system in glycerol fed-batch fermentation.

Keywords: automatic control system; fed-batch culture; pH logic control; supervisory computer control system

1. Introduction

1,3-Propanediol (1,3-PD) is one of bulk chemicals, which has been applied and used widely. Microbial production of 1,3-PD from glucose and/or glycerol has attracted much attention recently (Sun, Song, Sun, & Xiu, 2008; Zhao, Chen, & Yao, 2006). In recent years, there has been a growing interest in microbial production of 1,3-PD throughout the world because of its lower cost, higher production and no pollution (Biebl, Menzel, Zeng, & Deckwer, 1999; Xiu, Zeng, & An, 1999). There are three typical cultures for microbial fermentation of glycerol, i.e. batch, continuous and fed-batch cultures (Menzel, Zeng, & Deckwer, 1997). After a great deal of research on this biocconversion process of 1,3-PD has been made, including experimental investigation, modelling or parameter identification of this complex bioprocess (Yuan et al., 2014; Wang, 2012), and metabolic flux or metabolic pathway analysis (Wang, 2013), researches which focus on how to optimize the process become more and more valuable. Zhu, Yuan, Wang, Feng, and Xiu (2013) applied \( \mu \) synthesis method to deal with oscillatory behaviour in microbial continuous culture the biotechnology processes. Vector measure as controls for explicit nonlinear impulsive system of fed-batch culture has been discussed in Wang, Feng, and Xiu (2009). In this paper, we will focus on the fed-batch culture due to its high 1,3-PD concentration and productivity (Cheng et al., 2007).

During the fed-batch fermentation, glycerol and alkali are discontinuously added to the reactor so as to keep the substrate concentration and the pH at the desired levels. In the laboratory, on/off control of glycerol pump is determined by a pre-assigned times sequence. The addition of alkali can be coupled with the inlet flow of glycerol at a fixed ratio (Liu, Wang, Zhang, & Xiu, 2000) or determined by pH logic controller (Cheng, Sun, & Liu, 2004; Hao, Lin, Zheng, Sun, & Liu, 2008). In the former case, both the additions of glycerol and alkali are open-loop inputs, whereas the addition of alkali in the latter case is logically controlled.

It is hard to implement the fed-batch fermentation automatically because of the nonlinearity in cell growth and thus unable to achieve in real-time measurement of glycerol concentration. As a result, it is difficult to control glycerol concentration in a proper range, which leads to substrate inhibition to the cell growth and the productivity of 1,3-PD. At present, experimental researchers have to carry out several fermentations with various feeding strategies to get a reasonable one. The disadvantages of this methodology are that the operation is greatly dependent on human beings and there always exist great differences because of lots of uncertain factors. Moreover, consideration the expensive cost, it is impossible to carry out a number of experiments for various glycerol feeding strategies to obtain the optimal one. Therefore, mathematical modelling and optimal control for this microbial process become necessary.
In a recent study, Liu, Gong, and Feng (2009) proposed hybrid dynamical systems to describe the fed-batch culture with coupled feed of glycerol and alkali and explored its parameter identification and optimal control. Based on this, we have designed an automatic system to control such a process (Jiang, Ye, Feng, & Xiu, 2010). The strategy in fed-batch culture with coupled feed of glycerol and alkali is based on the hypothesis that the formation rate of acid by-products is positively related to the consumption rate of substrate. Previous experimental study also revealed that there exists functional relationship between the consumptions rate of the substrate and alkali (Tan, Wu, Li, & Wang, 2008), but this rate should not be constant in different fermentation stages. So controlling the pH by a logic controller should be much more precise than coupling it with the glycerol feed. For this reason, Ye, Feng, Yin, and Xiu (2011) further studied mathematical modelling of the fed-batch culture with glycerol open-loop inputs but pH logic control.

In this paper, by applying the optimal control algorithm proposed by Liu et al. (2009) on the model of Ye et al. (2011), we obtain the optimal control strategy for the fed-batch culture with open-loop glycerol input and pH logic control. Based on the above process, we design an automatic control system equipped with the existing fermentor, which is achieved by using programmable logic controller (PLC) to control the glycerol pump, alkali pump, drain pump, anti-foaming pump, the solenoid valve for water and electric heater. Additionally, an upper computer is designed to control the fermentation process globally, including writing the optimal control strategy, setting control parameters and so on. As a result, we provide the possibility of automatic control of 1,3-PD production in fed-batch culture.

The remainder of this paper is organized as follows: Section 2 introduces mathematically theoretical analysis for automatic control of glycerol fed-batch culture with pH logic control. Section 3 designs a control system to implement the optimal control strategy automatically. Conclusions are presented at the end of this paper.

2. Materials and methods

Klebsiella pneumoniae (DSM2026), which was purchased from the German Collection of Microorganisms and Cell Cultures, was used in this study. The preculture medium contained: \((\text{NH}_4)_2\text{SO}_4 2 \text{~g} / \text{L}, \text{K}_2\text{HPO}_4 3.4 \text{~g} / \text{L}, \text{KH}_2\text{PO}_4 1.3 \text{~g} / \text{L}, \text{MgSO}_4 0.2 \text{~g} / \text{L}, \text{yeast} \text{extract} 1 \text{~g} / \text{L}, \text{glycerol} 30 \text{~g} / \text{L}\). Preculture was carried out in 250-mL Xasks Willed with 50 mL medium. Flasks were incubated in a rotary shaker at 37° and 120 rpm for 14 h. During fermentation and culture, 50 mL of preculture was inoculated into a 5-L bio-reactor (BIOSTAT-B B.Braun Germany) with the working volume of 4 L. The fermentation was kept steadily at pH 6.8 and 37°. Medium used here was a chemically defined medium as described by Menzel et al. (1997). 40% (w/w) NaOH was used to adjust pH. Air feeding and rotation rate were 2 L/min and 300 rpm, respectively.

The biomass concentration was measured as optical density at 650 nm or dry weight. The determination of products (1,3-PD, ethanol and acetic acid) was carried out with a gas chromatograph (Shimazu GC-14B, FID-detector, \(2 \text{~mm} \times 0.5 \text{~mm}\) glass column packed with Chromosorb 101 and operated with He as carrier gas at flow rate of 40 mL/min, detector temperature of 220° and column temperature of 130°). Glycerol was assayed by a modified titration (Wang, Xiu, Liu, & Fan, 2001), in which glycerol was oxidized by sodium periodate to form formic acid, then sodium hydroxide was used to titrate formic acid produced.

3. Mathematical theory for automatic control of glycerol fed-batch culture with pH logic control

The fed-batch fermentation of glycerol with substrate feedforward and pH logic controls can be briefly described as follows. The process begins with batch fermentation, then glycerol and alkali are discontinuously poured into the reactor at constant flow rates in order that the concentration of glycerol keeps in a proper range and the pH of the solution in the desirable level (NaOH is used to neutralize the formed acids). The inputs of glycerol and alkali are determined by a pre-assigned time sequence and a pH logic controller, respectively.

According to the factual experiments, we can divided the fed-batch fermentation process into four different modes (indexed by Modes 0, 1, 2, 3) according to the on/off states of the pumps of glycerol and alkali. For details, see Ye et al. (2011).

Let \(C_m\) (mmol/L) and \(q\) (mmol/L) be the concentrations of glycerol and NaOH in the feed medium, respectively. Let \(F_{Gj}^0\) and \(F_{Gj}^0\), \(j \in I_3 = \{0, 1, 2, 3\}\) be the flow rates of alkali and glycerol, respectively, which take discrete values from finite sets \(S_1 := \{0, v_1\}\) and \(S_2 := \{0, v_2\}\), where \(v_1 > 0\) (L/h) and \(v_2 > 0\) (L/h) are constant feeding flow rates of alkali and glycerol. Let \(x = (x_1, x_2, \ldots, x_T)^T\), the components of which represent the concentrations of biomass (g/L), glycerol (mmol/L), 1,3-PD (mmol/L), acetic acid (mmol/L), ethanol (mmol/L), NaOH (mmol/L) in the reactor and the volume of solution (L), respectively. According to the factual experiments, we make the following assumptions.

(H1) The concentrations of reactants are uniform in reactor, while time delay and nonuniform space distribution are ignored.

Under the assumptions (H1), the Mode \(j\) \((j = 0, 1, 2, 3)\) of the fed-batch process can be described by

\[
\dot{x} = f^j(x, k), \tag{1}
\]

where \(k\) represents the kinetic parameter vector. The right-hand side of Equation (1) is of the form \(f^j(x, k) = \)

\[
\sum_{j=0}^{3} \sum_{k=1}^{4} k_{jk} \phi(x, k)
\]
The biological meanings of the parameters $\gamma$, $K_a$, $pK_w$, $K_w^{-}$, and $\epsilon_0$ in Equation (7) can be referred to Ye et al. (2011). Assume that the pH is expected to be in $[pH_{\text{min}}, pH_{\text{max}}]$. Once the value of the pH deceases to its allowable lower bound $pH_{\text{min}}$, the alkali pump will be forced to open and the alkali will be added to neutralize the acids in the solution; as long as the pH value reaches its allowable upper bound $pH_{\text{max}}$, the alkali pump will be shut down. The above processes are repeated until the end of the fermentation. In other words, to ensure that the pH is restricted in its admissible range, the following two inequalities must be satisfied during the entire fermentation time.

\begin{align}
h_0(x(t)) &:= pH_{\text{max}} - y_{\text{ph}}(x(t)) \geq 0, \quad (8) \\
h_1(x(t)) &:= y_{\text{ph}}(x(t)) - pH_{\text{min}} \geq 0. \quad (9)
\end{align}

Based on the above treatment, the process of fed-batch fermentation with open-loop glycerol input and pH logic control is described by the following HDS according to the previous work (Ye et al., 2011):

\[ \dot{x}(t) = f^{(\ast)}(x(t), k), \quad \bar{x}(t) = \bar{y}(x(t), k, \sigma) \text{ if } \xi(t) \in \text{Dom} F, \]

\[ \dot{\theta}(t) = \frac{1}{\sigma_{i,0}} \text{ if } \xi(t) \in \text{Dom} F, \]

\[ x(t+1) = x(t), \quad \bar{x}(t) = \bar{y}(x(t), k, \sigma) \text{ if } \xi(t) \in \text{Dom} F, \]

\[ \bar{y}(x(t), k, \sigma) := (f^{(\ast)}(x(t), k), \sigma), \quad \bar{y}(x(t), k, \sigma) := (f^{(\ast)}(x(t), k), \sigma) \in \mathbb{R}^n \times \mathbb{R}. \]

\[ x(t+1) = x(t), \quad \bar{x}(t) = \bar{y}(x(t), k, \sigma) \text{ if } \xi(t) \in \text{Dom} F, \]

\[ \theta(t+1) = \theta(t), \quad \bar{y}(x(t), k, \sigma) := (f^{(\ast)}(x(t), k), \sigma) \in \mathbb{R}^n \times \mathbb{R}. \]

The biological meanings of the parameters $\gamma$, $K_a$, $pK_w$, $K_w^{-}$, and $\epsilon_0$ in Equation (7) can be referred to Ye et al. (2011). Assume that the pH is expected to be in $[pH_{\text{min}}, pH_{\text{max}}]$. Once the value of the pH deceases to its allowable lower bound $pH_{\text{min}}$, the alkali pump will be forced to open and the alkali will be added to neutralize the acids in the solution; as long as the pH value reaches its allowable upper bound $pH_{\text{max}}$, the alkali pump will be shut down. The above processes are repeated until the end of the fermentation. In other words, to ensure that the pH is restricted in its admissible range, the following two inequalities must be satisfied during the entire fermentation time.

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Based on the above treatment, the process of fed-batch fermentation with open-loop glycerol input and pH logic control is described by the following HDS according to the previous work (Ye et al., 2011):

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\[ x(t+1) = x(t), \quad \bar{x}(t) = \bar{y}(x(t), k, \sigma) \text{ if } \xi(t) \in \text{Dom} F, \]

\[ \theta(t+1) = \theta(t), \quad \bar{y}(x(t), k, \sigma) := (f^{(\ast)}(x(t), k), \sigma) \in \mathbb{R}^n \times \mathbb{R}. \]

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\[ \theta(t+1) = \theta(t), \quad \bar{y}(x(t), k, \sigma) := (f^{(\ast)}(x(t), k), \sigma) \in \mathbb{R}^n \times \mathbb{R}. \]
where $\theta_i$ and $\bar{\theta}_i$ are empirical values. The set of solutions to the system (10), $S_0$, is defined as

$$S_0 := \{ x(\cdot; \theta) | x(\cdot; \theta) \text{ is a solution to the system (10)} \text{ with } \theta \in \Theta \}.$$ 

There exist critical concentrations, outside which cells cease to grow, of biomass, glycerol, 1,3-PD, acetate and ethanol. Hence, it is biologically meaningful to restrict the concentrations of biomass, glycerol, products, NaOH and the volume of culture fluid in a set $W$ given by

$$W := \prod_{i=1}^{7} [x_i, x_i^*].$$

Correspondingly, we denote the set of the admissible solutions by

$$S := \{ x(\cdot; \theta) \in S_0 | x(t; \theta) \in W, \forall t \in [0, T] \},$$

and the set of feasible decision vectors by

$$\Theta_F := \{ \theta \in \Theta | x(\cdot; \theta) \in S \}.$$ 

Then, optimizing the feeding instants of glycerol so as to maximize the concentration of 1,3-PD at the terminal time is formulated as follows:

$$\text{(OCP)} \quad \min \quad J(\theta) := -x_3(T; \theta),$$

$$\text{s.t.} \quad \theta \in \Theta_F.$$  

(12)

The problem (OCP) is a special case of the problem (OSCP) in Liu et al. (2009) with constant feeding rate of glycerol. Therefore, the algorithm constructed in Liu et al. (2009) can be directly applied to solve (OCP). For convenience, we denote the optimal decision vector by $\theta^*$. 

4. Automatic control design based on optimal control strategy

On the basis of the optimal control strategy in the previous section, we shall design an automatic control system for the fed-batch culture of 1,3-PD production with substrate feed-forward and pH value feedback controls.

A schematic diagram of the microbial fermentation process with the instrumentation installed is shown in Figure 1, which contains a glycerol pump, a foam pump, a drain pump, one alkali pump, a glycerol pot, a reactor, an alkali pot and one control box. The solid- and dashed lines in Figure 1 denote the liquid process and the path of electrical signal, respectively.

In consideration of the characteristics of the zymolysis technics in microbial production of 1,3-PD, the design projects of electrical diagram, PLC and monitoring system will be discussed.

4.1. The electrical design

The electrical design mainly includes control principle drawing, PLC wiring diagram and electric control panel layout, which are designed according to the national design specifications (Code for design of low voltage electrical installation; Code for design of electric distribution of general purpose utilization equipment).

The control principle drawing is shown in Figure 2. As shown in Figures 1 and 2, the fed-batch culture of 1,3-PD production involves the control of temperature, pH value, on/off switches of pumps (including glycerol pump, alkali
Figure 2. Control principle drawing.

pump, drain pump and anti-foaming pump) and the liquid level. A micro-circuit breaker is used as the power supply switch. AC contactor and thermal relay are adapted as control elements to every pump, which are designed as hand/auto knob control to make the control system more flexible.

PLC wiring diagram includes wiring of three I/O control units: digital input unit, digital output unit and analog input unit. In order to prevent input module and output module from being damaged in case of wrong wiring, the power supply (24V DC) for the PLC output module is separated from that for the input module. Considering the load capacity of PLC output ports, intermediate relays are added to the PLC output side, which is also helpful for improving the reliability of the control system.

In the design of the control box panel, the following two factors should be considered: providing suitable space for heat rejection and optimal arrangement of the elements.

4.2. PLC program design

We adopt PLC as the controller due to its superiority in data processing, communicating and networking and advantages of high reliability, abundant I/O port modules, modularized structure, easy installation and convenient service (Li, 2009). The control points of the PLC are designed according to the national design handbook (Handbook of industrial and civil power distribution, 2005). In this system, there are 14 digital inputs (12 signals from glycerol pump, alkali pump, the anti-foam pump and drain pump, each of which contain the signals of automation, running, overload, and 2 signals for the statuses of foam and water solenoid valve) and 6 digital outputs (controls of glycerol pump, alkali pump, anti-foam pump, drain pump, electric heater and the water solenoid valve). There are three analog signals: temperature sensor, pH value transmitter and liquid level transmitter. PLC is configured according to the statistics of the control points as shown in Figure 3. CPU with 24 points
(16 digital inputs and 8 digital outputs) and one analogical unit with 4 inputs for PLC is selected.

The four pumps, water solenoid valve and electrical heater are all controlled by a programme. All pumps are equipped with frequency inverters and work at a pre-assigned speed.

The control flow graph of fed-batch culture and pH value is shown in Figure 4. Herein, the control of glycerol feed is determined by the input of the optimal control strategy $\theta^*$ obtained in the previous section, whereas the pH value is controlled by alkali feed through a pH transmitter. Temperature is controlled by a solenoid valve and an electrical heater. The foaming single is measured by an electric pole installed on the top of the fermenter cover: when the formed foam contacts the bottom of the electric pole, an electric signal is produced in the measuring circuit, which imposes the anti-foam pump to run. The signal of the liquid level is transmitted by a liquid level transmitter.

### 4.3. Supervisory computer control system design

A computer is used as the management control platform in the central control room, through which the fermenter system is managed globally. The communication of the system is based on a wide area network with TCP/IP. The design of the display pictures includes six parts: main display window, control window, setting window, faults and alarms, real-time data report and history trends. The status of all equipments, analog values of the system and the course of the experiment are displayed on the main display window, which is the first picture that users open. As long as any buttons of equipments is clicked from the main display window, the control window is open instantly. This window is provided with functions of online manual control of equipments and start–stop control of the system. Type the correct login name and password in the login dialog field, the setting window is allowed to access to. On the setting window, operator can set duration, control strategy, control cycle for the fed-batch fermentation and the desired range for the pH value. The faults and alarms window monitor inputs and outputs, and sends alarm notifications to the operator. This window displays the most recent alarm notification first and many other alarm notifications on record as well. Real-time data report is used to display the real-time data values. The history trends display a graph of up to eight variables, and are used to record data and monitor the history operation of the system, making the system easy to be managed and analysed.

Kingview configuration software is applied to communicate with the PLC and provide a friendly human and machine interface for the user. The software design includes two parts: one is using Kingview software to program for upper computer to display the process of zymolysis techniques and data collected by PLC, the other is using PLC program software to program for the lower computer to carry out autocontrol. The communication linkage of hardware between the computer and the PLC is realized by RS232 cable. The settings of the communication parameter...
are as follows: baud rate: 19200bps, length of data bit: 8, length of stop bit: 1, parity check bit: odd. The communication parameter settings of the PLC should be consistent with that of the Kingview software. When configuring the PLC network, the address setting of the main linked unit of the PLC must be the same as that of Kingview software. The applicable range of address is from 0 to 31 defined by the Kingview software.

5. Discussions and conclusions
The proposed control system in this paper has the following advantages. Firstly, the design is reasonable because it is based on the optimal control strategy from the mathematically theoretical analysis. Secondly, the pH value is kept within the desired range more accurately by the closed-loop controller. Thirdly, the designed system provides a possibility of automatic control of microbial fed-batch production of 1,3-PD. Finally, the design has the advantages of higher equipment utilization ratio, lower running cost and convenient equipment management.

The simulator has been completed and works well so far. The optimized control strategy obtained in this work is much closed to that in Liu et al’s work (Liu et al., 2009). And the results are observed that the theoretical concentration of 1,3-PD can be increased considerably. Experiments for this control strategy under different manufacturing environments will be done gradually in our plan.

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