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## Modeling and identification of microbial batch fermentation using fuzzy expert system

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### ABSTRACT

The bioconversion of glycerol to 1,3-Propanediol by *Klebsiella pneumoniae* (*K. pneumoniae*) is a complex bioprocess. Because the transport mechanism of glycerol across cell membrane of *K. pneumoniae* was still not adequately understood, it is difficult to formulate reliable mathematical models. In this study, we aim to explore a novel model for describing the process of glycerol batch fermentation. This is achieved by a hybrid of a fuzzy expert system with a physical model framework. Some important properties of the proposed system are then discussed. To determine the model parameters, we establish a parameter identification model with the relative error of experimental data and simulating results as performance index. An optimization algorithm is developed to solve the identification model on the basis of constrain transcription and smoothing approximation techniques. Finally, the numerical simulations show the validity of the proposed model and the effectiveness of the optimization algorithm.

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### 1. Introduction

1,3-Propanediol (1,3-PD) as an important chemical product has numerous applications in medicines, cosmetics, lubricants, food and polymers [1]. Its microbial production is recently paid attention to throughout the world for its high production, low cost and no pollution [1–4]. Glycerol can be converted to 1,3-PD by several microorganisms [1]. Among all kinds of microbial productions of 1, 3-PD, glycerol bioconversion to 1,3-PD by *Klebsiella pneumoniae* (*K. pneumoniae*) has been widely investigated since the 1980s due to its high productivity [4].

A number of mathematical models have been developed to describe the fermentation process of glycerol by *K. pneumoniae*. In 1995, Zeng et al. proposed firstly a kinetic model to describe the fermentation process of glycerol [2]. The phenomena and characteristic of oscillation and hysteresis were then studied in [1,5]. In 2000, the model was improved by Xiu et al. [4], so that it not only can describe substrate consumption and products formation in a large range of glycerol concentrations in the feed medium, but also can predict the occurrence of multiplicity in this bioprocess. Thereafter, based on this model, parameter identification, system stability and optimal control of glycerol fermentation in continuous and batch culture have been extensively discussed [6,7]. For the fed-batch fermentation process of glycerol, nonlinear impulsive dynamical systems have been extensively investigated [8–11]. Taking the feeding process of glycerol as a time-continuous process, Wang proposed a novel hybrid system to describe the fed-batch fermentation process [12]. Subsequently, the properties, parameter identification problem and optimal control problem for the system have been investigated [13]. Nevertheless, some important intra-

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## Nomenclature

$K_1^*$	Monod saturation constant for substrate ( $\text{mmol L}^{-1}$ )
$K_2^*, K_4^*$	saturation constants for substrate and product in kinetic equations with excess terms ( $\text{mmol L}^{-1}$ )
$K_7^*, K_8^*$	inhibitor constant for 3-HPA to the enzyme GDHt and PDOR ( $\text{mmol L}^{-1}$ )
$Y_2, Y_4, Y_5$	maximum growth yield ( $\text{g mmol}^{-1}$ ) and product yield ( $\text{mmol g}^{-1}$ )
$\mu, \Delta q_1$	specific and maximum specific growth rate ( $\text{h}^{-1}$ )
$m_2, m_4, m_5$	maintenance term of substrate consumption and product formation under substrate-limited conditions ( $\text{mmol g}^{-1}\text{h}^{-1}$ )
$\Delta q_2, \Delta q_4$	maximum increment of substrate consumption rate and product formation rate under substrate-sufficient conditions ( $\text{mmol g}^{-1}\text{h}^{-1}$ )
$x_1(t)$	biomass concentration at time $t$ ( $\text{g L}^{-1}$ )
$x_2(t)$	extracellular glycerol concentration ( $\text{mmol L}^{-1}$ )
$x_3(t)$	extracellular 1,3-PD concentration ( $\text{mmol L}^{-1}$ )
$x_4(t)$	extracellular acetate concentration ( $\text{mmol L}^{-1}$ )
$x_5(t)$	extracellular ethanol concentration ( $\text{mmol L}^{-1}$ )
$x_6(t)$	intracellular glycerol concentration ( $\text{mmol L}^{-1}$ )
$x_7(t)$	intracellular 3-HPA concentration ( $\text{mmol L}^{-1}$ )
$x_8(t)$	intracellular 1,3-PD concentration ( $\text{mmol L}^{-1}$ )

cellular substances were not taken into consideration in the above models. In 2008, Sun et al. [14] firstly took three intracellular substances in reductive pathways into consideration and constructed a nonlinear enzyme-catalytic dynamical system to describe the continuous and batch fermentations of glycerol, in which it was assumed that glycerol passes the cell membrane of *K. pneumoniae* by passive diffusion coupling with active transport and 1,3-PD is transported by passive diffusion. On the basis of the literature [14], Ye et al. [15], Wang et al. [16] and Zhai and Yea [17], in consideration of three possible transport mechanisms (i.e., active transport, passive diffusion, active transport together with passive diffusion) for glycerol and 1,3-PD, constructed corresponding dynamical systems, respectively, and then inferred the most reasonable metabolic mechanism on the basis of a criterion that the most robust dynamical system is the most reasonable one.

However, to our best knowledge, the transport mechanisms of glycerol across cell membrane of *K. pneumoniae* were still not completely observed in the laboratory. Richery and Lin [18], Heller et al. [19], by comparing transport mechanisms of glycerol across cell membrane of *Escherichia coli* with that of *K. pneumoniae*, draw the heuristical knowledge that glycerol could pass cell membrane of *K. pneumoniae* by passive diffusion at high glycerol concentration and by active transport significantly only when the concentration of glycerol is at low concentration. In the previous literature [15–17], only a unique deterministic transport mechanism was assigned to the substrate glycerol in the so-call robustness dynamical system. So these models cannot describe the above heuristical knowledge in glycerol transport mechanism.

There are several different ways of representing heuristics knowledge, where the most popular technique is fuzzy logic [20]. Since the classical paper [21] of L.A. Zadeh, fuzzy system has gained recognition and was intensively applied in mathematics and computer sciences [22,23]. By combining black box techniques (e.g. fuzzy system and neural network) with a physical model framework, hybrid models can be obtained. Such models have the advantages of high level of interpretability and the ability to deal with complex nonlinear behavior, and have been widely used in biological modeling studies, especially in the case that the mechanisms ruling the biological processes are not adequately understood [24,25].

In this paper, we study the modeling of glycerol bioconversion to 1,3-PD in batch culture. Based on qualitative heuristic knowledge from biochemists, a hybrid model is constructed to describe the batch culture by introducing a fuzzy expert system into the enzyme-catalytic dynamical system. Some important properties of the proposed system are then discussed. To determine the parameters of the proposed system, a parameter identification model with the relative error of experimental data and simulating results as performance index is established. To solve the identification model, an optimization algorithm is developed on the basis of constrain transcription and smoothing approximation techniques. Numerical simulations indicates that the proposed hybrid model is proper.

The rest of this paper is organized as follows. In Section 2, a hybrid model of batch culture is established. In Section 3, some important properties of nonlinear dynamical systems are investigated. In Section 4, we propose a parameter identification model for the system and construct a feasible algorithm to solve it. Conclusions are presented at the end of this paper.

## 2. Nonlinear dynamical system of batch culture

The fermentation of glycerol covers both extracellular and intracellular environments which are linked by the transports of substrate and products across cell membranes. During glycerol metabolism, glycerol is firstly transported to the intracellular environment from the extracellular across membrane, and then is further catabolized, reactions catalyzed by enzymes, to generate intermediates and final product 1,3-PD, finally, intracellular 1,3-PD is transported to the fermentative broth as

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