

# Combined effects of temperature and pH on biohydrogen production by anaerobic digested sludge

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#### ARTICLE INFO

Article history: Received 21 December 2009 Received in revised form 30 May 2011 Accepted 2 June 2011 Available online 29 June 2011

Keywords: Hydrogen production Biohydrogen Full factorial design Kinetic model Anaerobic sludge

## ABSTRACT

A full factorial design was conducted to investigate the combined effects of temperatures and initial pH on fermentative hydrogen production by mixed cultures in batch tests. The experimental results showed that the modified Logistic model can be used to describe the progress of cumulative hydrogen production in the batch tests of this study. The modified Ratkowsky model can be used to describe the combined effects of the temperatures and initial pH on the substrate degradation efficiency, hydrogen yield and average hydrogen production rate. The temperatures and initial pH had interactive impact on fermentative hydrogen yield and the maximum substrate degradation efficiency, the maximum hydrogen yield and the maximum average hydrogen production rate was predicted at the temperature of 37.8 °C and the initial pH of 7.1, 37.4 °C and 6.9, and 38.2 °C and 7.2, respectively. In general, the optimal temperature for the fermentative hydrogen production was around 37.8 °C and the optimal initial pH for the fermentative hydrogen production was around 7.1.

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## 1. Introduction

Hydrogen is one of alternative energy source that are environmentally friendly and renewable. It produces only water, when it is combusted as a fuel or converted to electricity. Since fermentative hydrogen production can be carried out at ambient temperature and pressure, it is less energy-intensive than chemical and electrochemical hydrogen production processes. In addition, it is of great significant to produce hydrogen from organic wastes by fermentative hydrogen production, because it can not only treat the organic wastes, but also generate very clean fuel hydrogen. Therefore it has been received increasing attention in recent years [1–4].

Fermentative hydrogen production processes using mixed cultures are more practical than those using pure cultures, because the former are simpler to operate and easier to control, and may have a broader source of feedstock, thus mixed cultures are preferred for waste treatment. Yet in a fermentative hydrogen production process using mixed cultures, the hydrogen produced by hydrogen-producing bacteria can be consumed by hydrogen-consuming bacteria; thus, in order to harness hydrogen from a fermentative hydrogen production process, the seed sludge needs to be pretreated to suppress as much hydrogen-consuming bacterial activity as possible while still preserving the activity of the hydrogen-producing bacteria. It has been shown that heat-shock is simple and effective to suppress hydrogen-consuming bacteria and enrich hydrogenproducing bacteria [2].

Many factors such as temperatures and initial pH can influence the fermentative hydrogen production, because they can affect the activity of hydrogen-producing bacteria by influencing the activity of some essential enzymes such as hydrogenases for fermentative hydrogen production [5]. It has been demonstrated by various one-factor-at-a-time experiments that in an appropriate range, increasing temperatures and initial pH respectively could increase the ability of

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<sup>0961-9534/\$ —</sup> see front matter @ 2011 Elsevier Ltd. All rights reserved. doi:10.1016/j.biombioe.2011.06.016

hydrogen-producing bacteria to produce hydrogen, but temperatures and initial pH at much higher levels could decrease it with increasing levels [6–13].

Statistical design for experiments can be used for process characterization, optimization and modeling. It has been widely used for improving product performance, process capability and yield. In a statistical design, the levels for factors involved in an experiment are simultaneously varied. Thus, a lot of information can be taken with minimum runs of experiment. The statistical design for experiments in which the effects of more than one factor on response are investigated is known as full factorial design. Its most important advantage is that, not only the effects of individual factors, but the interactive effects of two or more factors can also be investigated, which is not possible in a classical one-factor-ata-time design [14].

Since a full factorial design has advantages over the onefactor-at-a-time method for investigating the effects of more than one factor on a process, it was adopted in this study to investigate the combined effects of temperatures and initial pH on fermentative hydrogen production by mixed cultures in batch tests, with the purpose of obtaining the optimal temperature and initial pH for fermentative hydrogen production.

## 2. Materials and methods

## 2.1. Seed sludge

The digested sludge collected from a primary anaerobic digester at Beijing Gaobeidian Sewage Treatment Plant (China) was used as the seed sludge. The volatile suspended solid (VSS) of the sludge was 11.8 g/L. Since previous studies showed that the biogas produced by the sludge pretreated by heat-shock contained only hydrogen and carbon dioxide, without detectable methane [1,2], heat-shock was used in this study to enrich hydrogen-producing bacteria by heating the seed sludge at 100 °C for 15 min.

## 2.2. Experimental design and procedures

A full factorial design was used to design the experiment in this study. The substrate degradation efficiency, hydrogen yield and average hydrogen production rate were chosen as the response variables, while temperatures and initial pH were chosen as two independent variables. The experimental design is shown in Table 1.

According to the experimental design in Table 1, batch tests were conducted in 150 mL glass bottles. One liter of the nutrient solution contained NaHCO<sub>3</sub>, 40,000 mg; NH<sub>4</sub>Cl, 5000 mg; NaH<sub>2</sub>PO<sub>4</sub>·2H<sub>2</sub>O, 5000 mg; K<sub>2</sub>HPO<sub>4</sub>·3H<sub>2</sub>O, 5000 mg; FeSO<sub>4</sub>·7H<sub>2</sub>O 15,000 mg; MgCl<sub>2</sub>·6H<sub>2</sub>O 85 mg; NiCl<sub>2</sub>·6H<sub>2</sub>O 4 mg. Fifteen milliliters of the pretreated seed sludge, 10 mL of nutrient solution and 1 g of glucose were added to each glass bottle, respectively. And then the total working volume of the bottles was filled to 100 mL by de-ionized water. The initial pH of the mixed solution in each bottle was adjusted by 1 mol/L HCl or 1 mol/L NaOH. Each bottle was flushed with argon for 3 min to provide anaerobic condition, capped with a rubber

Table 1 – Experimental design and the corresponding	
experimental results.	

Run	Т (°С)	Initial pH	Substrate degradation efficiency (%)	Hydrogen yield (mL/g glucose)	Average hydrogen production rate (mL/h)
1	30.0	6.0	70.5	236.8	8.4
2	30.0	7.0	80.7	262.0	9.8
3	30.0	8.0	75.8	224.7	9.1
4	30.0	9.0	52.5	136.5	6.3
5	35.0	6.0	82.6	278.8	9.7
6	35.0	7.0	95.6	308.5	11.4
7	35.0	8.0	83.6	264.6	10.5
8	35.0	9.0	64.6	160.7	7.3
9	40.0	6.0	80.4	277.9	9.8
10	40.0	7.0	96.5	307.4	11.5
11	40.0	8.0	87.5	263.7	10.7
12	40.0	9.0	60.2	160.2	7.4
13	45.0	6.0	71.0	228.2	8.3
14	45.0	7.0	80.0	252.5	9.7
15	45.0	8.0	72.2	216.5	9.0
16	45.0	9.0	52.3	131.5	6.3

stopper, and placed in a reciprocal shaker (reciprocation: 150 strokes/min) at different temperatures. Each batch test was done three replicates.

#### 2.3. Analytical methods

The water displacement method was used to collect and measure the biogas produced. The fraction of hydrogen in the biogas was determined by a gas chromatograph (Model 122, Shanghai, China) equipped with a thermal conductivity detector (TCD) and a 2 m column packed with 5 Å molecular sieves. Helium was used as the carrying gas at a flow rate of 12 mL/min. The operating temperatures of the column, detector and injector were 40 °C, 80 °C and 50 °C, respectively. All gas production data reported were standardized to the standard temperature (0 °C) and atmospheric pressure (1 atm). The pH in the solution was measured by a pH meter (Model 526, Germany). The concentrations of glucose after reaction were determined by the DNS colorimetric method [15]. The volatile suspended solid (VSS) concentration was determined according to the standard methods [16].

### 2.4. Data analysis methods

The modified Logistic model (Eq. (1)) was used to describe the progress of cumulative hydrogen production in the batch tests of this study [4].

$$H = \frac{P}{1 + \exp[4R_m \cdot (\lambda - t)/P + 2]}$$
(1)

where *H* (mL) is the cumulative hydrogen production at the reaction time t (h), *P* (mL) is the hydrogen production potential,  $R_m$  (mL/h) is the maximum hydrogen production rate and  $\lambda$  (h) is the lag time.

In this study, the modified Logistic model was used to fit the cumulative hydrogen production data obtained from each Download English Version:

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