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Optimization of photosynthetic hydrogen production from acetate by *Rhodobacter sphaeroides* RV

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ABSTRACT

In this study, hydrogen production by *Rhodobacter sphaeroides* RV from acetate was investigated. Ammonium sulphate and sodium glutamate were used to study the effects of nitrogen sources on photosynthetic hydrogen production. The results showed the optimal concentrations for ammonium sulphate and sodium glutamate were in the range of 0.4–0.8 g/L. Orthogonal array design was applied to optimize the hydrogen-producing conditions of the concentrations of yeast, $FeSO_4$ and $NiCl_2$. The theoretical optimal condition for hydrogen production was as follow: yeast 0.1 g/L, $FeSO_4$ 100 mg/L and $NiCl_2$ 20 mg/L.

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1. Introduction

The energy gap gradually increased due to rapid depletion of fossil fuels. In addition, the consumption of fossil fuels induced environmental pollution and global warming [1]. So there is necessity to find clean, renewable energy sources that could replace fossil fuels in the future. Solar energy is the most abundant amongst several renewable energy sources, but with associated lower efficiency of converting solar energy into energy carriers [2]. The photosynthetic bacteria can use a wide part of the solar spectrum (400–950 nm), and utilize organic substrates (organic acids, sugars, etc.) to produce hydrogen [3–5]. Hydrogen gas is widely recognized as an ideal and clean energy carrier because of its cleanness, renewable character, and the highest gravimetric energy density (122 kJ g^{-1}) [6–8]. As compared to the conventional chemical and electrolytical routes to produce hydrogen [6,9], photosynthetic hydrogen production provides a promising ways because of combined wastewater treatment with energy recycling [10–12]. In addition, photosynthetic bacteria have high substrate conversion yield for transforming solar energy into hydrogen (12 mol H_2/mol glucose). The actual yields are much lower than the theoretical maximum value, which limits the large-scale production of hydrogen [13].

Several researchers have optimized culture parameters to improve photosynthetic hydrogen-producing yield, such as carbon source, nitrogen source, C/N ratio, temperature, light intensity as well as nutrient medium [3,4,10,13–16]. Nitrogen source is a crucial factor among several hydrogen-producing parameters. Nitrogen source can influence the activity of

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nitrogenase, while hydrogen production is associated mainly and completely with the action of nitrogenase [17,18]. A few reports show that metal ions can enhance hydrogen production for certain photosynthetic bacteria [19,20]. The reason is probably that metal ions are components of several enzymes, which are able to catalyse hydrogen evolution. Researchers have been trying to optimize photosynthetic hydrogen production, but high conversion efficiency of substrate has not been obtained.

Rhodobacter sphaeroides is a group of bacteria that can obtain energy through photosynthesis and has been applied into photosynthetic hydrogen production [11,16,20]. The objective of this study was to determine the effects of nitrogen source, yeast, ferrous and nickel concentrations on biohydrogen production with *R. sphaeroides* RV. The data obtained from this study are expected to provide basic and engineering data for the large-scale studies of photosynthetic hydrogen production.

2. Materials and methods

2.1. Inoculum, medium and analytical procedures

The inoculum of *R. sphaeroides* RV, the synthetic medium and the analytical procedures were the same as our previous work described in Ref. [21].

2.2. Experimental method in photo-fermentation

Hydrogen-production batch experiments for nitrogen source were performed in 40 mL glass column photobioreactor (working volume, 30 mL). Effects of yeast, ferrous and nickel concentrations on hydrogen production were conducted in serum bottles of 120 mL (working volume, 80 mL). The temperature was maintained at 32 °C. Light intensity was 220 W/m^2 at the outer surface of the reactor. The basic medium and additional carbon sources were used as the medium for photosynthetic hydrogen production. Required amount of sodium acetate was added to adjust concentration to 0.8 g/L. pH was adjusted to 7.0 with 4% sodium hydroxide and hydrochloric acid. Argon gas was used to create anaerobic conditions. The gas volume was calibrated to 25 °C and 760 mmHg. All the experiments were done in batch operation. Each experimental condition was carried out in triplicate. The experimental results noted were the averages (±standard deviation) of the values obtained in independent experiments conducted in triplicate. All chemicals used in the experiments were of AR grade.

2.3. Model analysis

The cumulative hydrogen production in the batch experiments followed the modified Gompertz equation [22]:

$$H = P\exp\left\{-\exp\left[\frac{R_m e}{P}(\lambda - t) + 1\right]\right\}$$

where *H* is the cumulative hydrogen production (mL), *P* is hydrogen production potential (mL), *Rm* is the maximum hydrogen production rate (mL/h), *e* is 2.71828, λ is the lag-phase time (h), and *t* is the incubation time (h). The corresponding values of *P*, *Rm* and λ for each batch were estimated using Origin 7.5, which is a scientific graphing and data analysis software.

3. Results and discussion

3.1. Effects of nitrogen sources on hydrogen production

Ammonium sulphate and sodium glutamate as nitrogen sources were used in the experiments, as they are the most widely used as nitrogen sources for hydrogen production. Batches were dosed with nitrogen source at 0, 0.4, 0.8, 1.2, 1.6, 2.0, 2.4, 2.8, 3.2 and 3.6 g/L. Fig. 1 shows the effects of ammonium sulphate concentrations on cumulative hydrogen production, OD_{660} (Fig. 1a), acetate consumption and nitrogen consumption efficiencies (Fig. 1b). Ammonium sulphate concentrations strongly affected hydrogen production. The cumulative hydrogen production were higher than others bottles when the concentrations of ammonium sulphate were within 0.4–1.6 g/L. The maximum cumulative hydrogen production (4.8 mL) was obtained at the ammonium sulphate

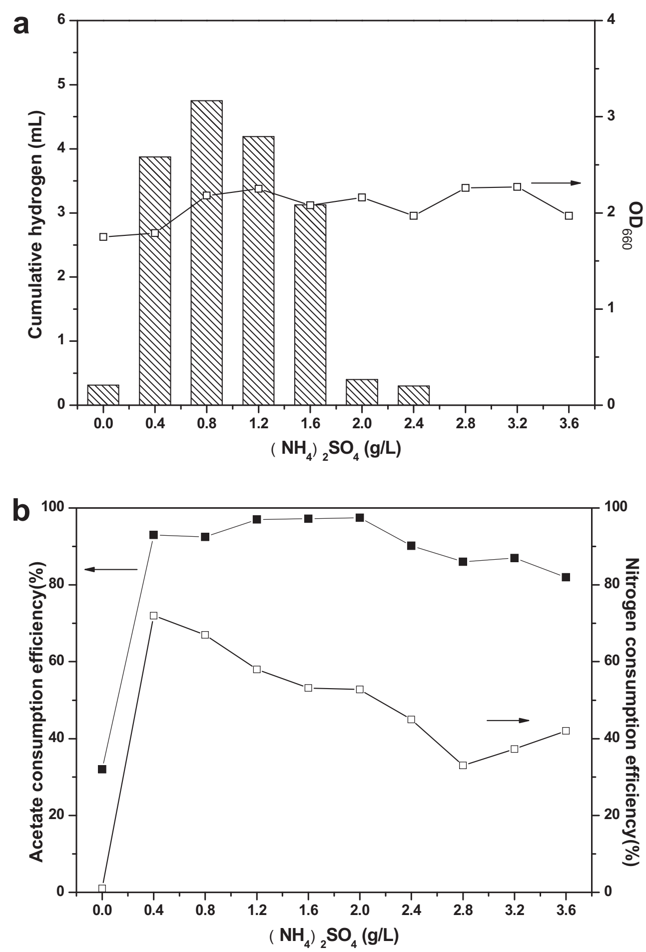

Fig. 1 – Effects of ammonium sulphate concentrations on cumulative hydrogen production, OD_{660} (a), acetate and nitrogen consumption efficiencies (b).

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