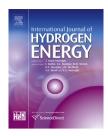
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Photosynthetic hydrogen production from enzyme-hydrolyzed micro-grinded maize straws

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ABSTRACT

Photosynthetic bacteria can produce hydrogen from various organic substrates in the presence of illumination energy. This study produced biohydrogen from maize straws by an enriched consortium via photo-fermentation pathway. The maize straws can be utilized by the consortium as substrate only after micro-grinding and enzyme hydrolysis pre-treatments. Batch tests at 30 g/L maize straw, pH 7.0, 30 °C, illumination intensity 2000 lx yielded 9.33 mol H₂/mol glucose with illumination energy conversion rate of 13.7% and substrate-energy conversion rate of 5.3%. Equivalently, the biohydrogen productivity is about 3 mmol/g maize straw. Reaction temperature can affect hydrogen production rate but only minimally affect the delayed time. The activation energy for biohydrogen production was estimated to be 5.65 kJ/mol.

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Introduction

Biohydrogen is clean energy produced from biomass [1,2]. Hydrogen production by photosynthetic bacteria was intensively studied [3–7]. Since the photo-fermentation process can produce hydrogen from organic substrates in the presence of illumination light, it was proposed to work following the dark-fermentation process for maximizing hydrogen yield [8–11].

Biomass needs hydrolysis for reducing high-molecular weight polymers to reduced sugars [12,13]. Physical,

chemical and biological pretreatments were proposed to enhance hydrolysis of biomass [14–17]. Micro-grinding is a process to force the feed material through a gap between a stationary surface and a moving surface with bursts and grooves to crush the cellulosic matters to micro-sized fragment by mechanical force [18,19]. Micro-grinding is a costeffective and efficient pretreatment for biomass disintegration; however, few studies on the use of micro-grinding for biomass pre-treatment to enhanced biohydrogen production are available.

This study disintegrated the structure of maize straws by micro-grinding followed by enzyme treatment to increase

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the reduced sugar, then the pre-treated straws were used as the substrate for photo-fermentative hydrogen production. The modified Gompertz model was used to describe experimental data with model parameters being estimated by data fitting.

Experimental

Materials

Maize straw was collected from suburb areas of Zhengzhou City, China, was dried at 55 °C for 72 h as the raw material. The dried material was micro-grinded by a LG-02 grinder (Hangming Co., Zhengzhou, China) at rotational speed 25,000 rpm, 0.6 kW for 300 s. The grinded powders were screened by 240–270 standard meshes to have particle size ranging 53–61 μ m.

The photosynthetic bacterial consortium was made by enriching the isolated strains F1, F5, F7, F11 (all photosynthetic purple non-sulfur bacteria), L6 (Chlorobiaceae), S7 and S9 (purple sulfur bacteria), according to the cultivation methods proposed previously [20]. These isolated were noted to have high biohydrogen yields from most organic acids and glucose while their mix could produce more hydrogen than individual isolate. The enrichment medium was composed of 0.2 g/L MgCl₂, 0.4 g/L NH₄Cl, 0.5 g/L K₂HPO₄, 2 g/L NaCl, 3.56 g/L sodium glutamate, 0.1 g/L yeast extract. The consortium was grown in this medium at 30 °C for 50 h to reach logarithmic growth phase.

The growth medium for hydrogen production test was composed of 0.2 g/L K₂HPO₄, 0.2 g/L MgSO₄, 1 g/L NH₄Cl, 2 g/L NaHCO₃, 2 g/L NaCl, 4 g/L CH₃COONa, 1 g/L yeast extract, 5 mL/ L growth factor (0.1 mg/L vitamin B₂, 0.1 mg/L biotin, 10 mg/L vitamin B₃, 10 mg/L 4-aminobenzoic acid), and 2 mL/L trace elements (0.05 mg/L CuSO₄·6H₂O, 5 mg/L FeCl₃·6H₂O, 0.05 mg/L MnCl₂·4H₂O, 1 mg/L ZnSO₄·7H₂O, 1 mg/L H₃BO₄, 0.5 mg/L Co(NO₃)₂·6H₂O).

Apparatus and tests

A double-layered vacuum glass flask with an effective volume of 400 mL was the photoreactor. Both sides of the flask were equipped with 30 W incandescent lamps for providing 2000 lx intensity to the reactor. The entire reactor and incandescent lamps were placed in the LRH-250-GS constant temperature incubator at prescribed temperature of 20-40 °C.

12 g grinded and hydrolyzed maize was placed to a few 500 mL flasks together with 320 mL of sodium citrate buffer (pH 4.8) and 0.6 g cellulase (CAS No. 9012-54-8; Solarbio Co., Beijing, China). The resulting suspension has solid concentration of 30 g/L. The suspension was shaken at 150 rpm at 50 °C for 48 h. Then the suspension was titrated to pH 7.0 by adding NaOH. The pretreated suspension was placed into the double-layered vacuum flask for biohydrogen test. Then the growth medium and the enriched consortium were added at 4:1 v/v to the reactor. The produced gas was collected from the reactor to pass through a 50% w/w NaOH solution to remove CO_2 . The gas production quantity for the purified gas was

measured by drainage method. The testing period was lasted for 168 h.

The solution pH was measured by pHS-2C pH meter (Jiangsu Zhengji Instruments, Jiangsu, China) and the illumination intensity at reactor side was measured by LX1010B intensity meter (Shinbou Sci., Shenzhen, China). The hydrogen concentration of purified gas was measured by a GC-16B gas chromatograph (PerkinElmer, MA, USA) equipped with 5A column and thermal conductivity detector (TCD) with 45 mL/min nitrogen as the carrier gas.

Results and discussion

Hydrogen production

Hydrogen production rates at 20–40 °C are shown in Fig. 1. The corresponding specific hydrogen production rates (dV/dt) are estimated by graphic differentiation and are shown in Fig. S1. Control tests revealed that the enriched bacterial consortium could not yield hydrogen from either the growth medium (Sec. "Materials") or the hydrogen production medium without pretreated maize straws. Independent control tests also revealed that the enriched consortium could not produce biohydrogen via photo-fermentation from maize without micro-grinding or enzyme hydrolysis. Restated, the hydrogen could be produced only from those maize straws after both grinding and enzymatic hydrolysis.

The hydrogen production rate was increased with time, peaked at hr 72, and then declined in further testing. The hydrogen production rate was rapidly increased as temperature was increased from 20 to 30 °C (from 6.5 to 26.4 mL/L-h), and then was slightly decreased when the temperature was further increased to 40 °C (to 21.2 mL/L-h). The optimal temperature for photosynthetic hydrogen production from grinded-enzyme-hydrolyzed maize straw by the tested consortium was 30 °C.

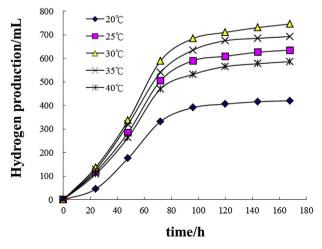


Fig. 1 – Cumulative hydrogen production with time at different reaction temperature. 30 g/L maize straw, pH 7.0, illumination intensity 2000 lx.

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