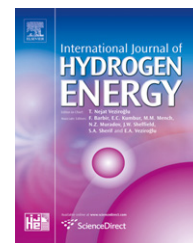


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Performance of a groove-type photobioreactor for hydrogen production by immobilized photosynthetic bacteria

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ABSTRACT

The biofilm technique has been proved to be an effective cell immobilization method for wastewater biodegradation but it has had restricted use in the field of photobiological H_2 production. In the present study, a groove-type photobioreactor was developed and it was shown that a groove structure with large specific surface area was beneficial to cell immobilization and biofilm formation of the photosynthetic bacteria on photobioreactor surface as well as light penetration. A series of experiments was carried out on continuous hydrogen production in the groove-type photobioreactor illuminated by monochromatic LED lights and the performance was investigated. The effects of light wavelength, light intensity, inlet glucose concentration, flow rate and initial substrate pH were studied and the results were compared with those obtained in a flat panel photobioreactor. The experimental results show that the optimum operational conditions for hydrogen production in the groove-type photobioreactor were: inlet glucose concentration 10 g/L, flow rate 60 mL/h, light intensity 6.75 W/m², light wavelength 590 nm and initial substrate pH 7.0. The maximum hydrogen production rate, H_2 yield and light conversion efficiency in the groove-type photobioreactor were 3.816 mmol/m²/h, 0.75 mol H_2 /molglucose and 3.8%, respectively, which were about 75% higher than those in the flat panel photobioreactor.

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1. Introduction

As a clean, sustainable and efficient energy carrier, hydrogen is considered to play a key role in future energy supplies and to be one of the expected ways to avoid problems caused by fossil fuels [1]. There are many ways to produce hydrogen nowadays but most are energy intensive, which makes hydrogen production expensive and contributes to further environmental contamination. Hydrogen generation via biological routes is expected to be one of main feasible approaches to hydrogen energy supply due to energy saving, high efficiency and pollution-free. Hydrogen can be produced by four types of microbes, including anaerobic fermentative bacteria, photosynthetic bacteria (PSB), cyanobacteria and

algae [2–7]. Among them, photosynthetic bacteria, which can utilize a wide range of the solar spectrum (400–950 nm) and degrade short-chain organic acids as electron donors to produce H_2 , are the most attractive candidate. Furthermore, this bio-energy process coupling with wastewater treatment and solar energy utilization on a large scale can be operated at ambient temperature and pressure. Thus, many researchers have focused their studies on PSB for hydrogen production.

It should be pointed out that one necessary premise of practical photobiological hydrogen production is to make the process operate continuously. Cell immobilization is a feasible technique to realize the continuous hydrogen production. Two immobilized-cell techniques, gel entrapment and biofilm, have been applied in biological hydrogen production.

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For gel entrapment, the main drawbacks are low hydrogen production rate, insufficient light supply, weak mechanical strength and poor stability for long-term operation [8–10]. The low hydrogen production can be attributed to high mass transfer resistance in a fixed bed bioreactor and the insufficient light supply is due to gel opacity [11–13]. Immobilized-cell systems with biofilm formation are considered as suitable means for continuous hydrogen production due to their advantages of high substrate conversion efficiencies, preferential retention of active microbial mass and avoidance of product inhibition. In general, the hydrogen production rate of a bioreactor with PSB immobilized on a solid matrix is higher than that with free-living cells [14–16]. However, technical barriers, such as poor mass transfer, insufficient immobilized biomass and low light conversion efficiency, will still limit the performance of photobioreactors. This is the motivation of the present study.

Bacterial adherence to the substratum is a critical step in biofilm formation. Investigations revealed that cells facing physical, chemical and biological threats from their environment prefer to adhere to the surfaces of channels or tubes [17,18]. As adherent cells grow and divide, physiological adaptations, including secretion of extracellular polymeric substance (EPS), are induced to create a protective matrix surrounding the cells. These dynamic communities can spread across surfaces and incorporate new planktonic cells to ultimately form a biofilm [19,20]. One can see that surface feature plays an important role in the biofilm formation. Literatures on the dependence of biofilm formulation on substratum materials and surface topography have revealed that rough or patterned surfaces can enrich cell adherence compared with smooth surfaces. Walker and Marsh [21] reviewed biofilm formulation and its role in microbial contamination of dental unit water systems (DUWS) with narrow-bore tubing and concluded that DUWS were susceptible to biofilm and encouraged biofilm growth. Carlén et al. [22], who studied surface characteristics and biofilm formation on unpolished and polished glass ionomer and composite resins for the purpose of tooth protection, showed that higher surface roughness led to more inorganic and better bacterial adherence. Whitehead and Verran [23] compared experimental results of cell retention on four stainless steel surfaces for the purpose of avoiding corrosion of stainless steel, and they found that cells were more evenly spread across titanium-coated surfaces than fine polished surfaces and cell numbers were higher. Meanwhile, surface topography influenced the pattern of cell retention. Scheuerman et al. [24] studied cell adhesion on a silicon coupon with etched grooves perpendicular to the flow direction. Their results showed that the maximum initial accumulation was at the bottom of the rough elements due to protection from shearing stress. Ginsburg and Karamanev [25] investigated the roughness effect of a graphite surface on the immobilization of *Acidithiobacillus ferrooxidans* and found that activated carbon fiber with the largest surface area per gram led to the maximum immobilized microorganisms. Hence, it can be expected that rough surfaces can definitely be beneficial to PSB immobilization and biomass enrichment in a photobioreactor, hence improving hydrogen production. However, little research has been devoted to the effect of surface

topography on the performance of photobioreactors with immobilized PSB cells.

In the present study, a novel photobiological reactor with grooved surface was developed for continuous hydrogen production by immobilized PSB. The performances with respect to hydrogen production rate, hydrogen yield and light conversion efficiency of the photobioreactor were investigated and the effects of operation conditions, including illumination wavelength and intensity, concentration, flow rate and pH value of the influent substrate, were discussed and compared with that of a photobioreactor with flat panel surface.

2. Materials and methods

2.1. Microorganism and medium

An indigenous photosynthetic bacterium cell identified as *Rhodospseudomonas palustris* CQK 01 was used for photoheterotrophic hydrogen production in the present study. The strain with steady and high hydrogen production capacity was isolated from local municipal sewage sludge [26]. The bacterium grew in a synthetic medium consisting of: $K_2HPO_4 \cdot 3H_2O$ (1.006 g/L), KH_2PO_4 (0.544 g/L), $MgSO_4 \cdot 7H_2O$ (0.2 g/L), $FeSO_4 \cdot 7H_2O$ (0.0417 g/L), $C_5H_8NNaO_4$ (0.5 g/L), $C_6H_{12}O_6 \cdot H_2O$ (9.9 g/L), NaCl (0.2 g/L), $CaCl_2$ (0.01 g/L), $(NH_4)_6Mo_7O_{24} \cdot 4H_2O$ (0.001 g/L), $ZnSO_4 \cdot 7H_2O$ (0.001 g/L), H_2NCONH_2 (1.677 g/L), yeast extract (1.0 g/L) and 1 mL growth factor solution (biotin, 1.0 g/L; pyridoxine hydrochloride, 1.0 g/L; riboflavin, 1.0 g/L; thiamine hydrochloride, 1.0 g/L; nicotinic acid, 1.0 g/L). Prior to inoculation, the cells were cultivated anaerobically in 150 mL culture medium at 30 °C for 48 h under illumination with a tungsten filament lamp at 30 W/m² until the strain was harvested in their exponential growth phase. The culture medium was autoclaved (20 min, 120 °C, 1.2 bar) before use. An anaerobic condition was created by argon gas. The initial pH value of the medium was adjusted to 7.0 by 0.1 M NaOH before incubation.

2.2. Photobioreactors

A novel groove-type photobioreactor, as shown in Fig. 1, was developed in the present study. It was a sealed vessel with working volume of 100(H) × 50(L) × 20(W) mm³ fabricated from polymethyl methacrylate (PMMA). PMMA was proved to be suitable for photobiological hydrogen production due to its transparency, chemical stability and ease of mechanical incising. Grooves (1 mm width and 1 mm depth) were etched by a mechanical incising technique on one vessel wall with 100 mm length and 50 mm width. Furthermore, a flat panel photobioreactor with the same volume as the groove-type one was fabricated and used as a comparison.

2.3. Experimental setup and conditions

Schematic details of the experimental setup are depicted in Fig. 2. The experimental system consisted of a biofilm photobioreactor, a light source, a peristaltic pump, a substrate medium flask, a liquid effluent flask, a gas-liquid separator,

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