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Preparation of a microalgal photoanode for hydrogen production by photo-bioelectrochemical water-splitting

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ABSTRACT

In this study, a microalga *Tetraselmis subcordiformis* (synonym: *Platymonas subcordiformis*)-based photoanode was prepared by a novel method developed in our lab. The optimal photocurrent density of microalgae photoanode, 37 μA/cm^2, was achieved under illumination of 145 μmol s^{-1} m^{-2} at anode potential of 0.5 V vs Ag|AgCl|sat. KCl, immobilized cell density of 2.08 × 10^6/cm^2 and BQ concentration of 300 μmol/L. The results of measurements showed that oxygen evolution peak, hydrogen evolution peak and photocurrent response were all synchronous to light impulse in a three-electrode system. It revealed that there occurred a process of photo-bioelectrochemical water-splitting. Hydrogen can be produced by the method. The investigation for whole photo-bioelectrochemical process also indicated that the electrons for hydrogen evolution had two sources, microalgal metabolic process in dark condition and photosynthetic water oxidation. The photo-hydrogen evolution was twice more than hydrogen evolution in dark condition.

1. Introduction

Oxygenic photosynthesis, biological water-oxidation, is considered as the most important process among all energy converting processes on earth [1]. The high-energy electrons produced from water oxidation in the photosystems could be potentially transferred to electrodes through exogenous mediators to generate a photocurrent. Some studies have been done to investigate photo-bioelectrochemical anodic reactions [2,3].

Photoelectrochemical (PEC) water-splitting, which was first demonstrated by Fujishima and Honda [4], provides us one particularly promising approach [5]. In the method, water oxidation (producing O_2) and hydrogen reduction (producing H_2) are spatially separated by having each process occur at a separate electrode. Meanwhile, an applied (electrical or

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chemical) bias is needed to compensate for insufficient PEC cell voltage and overcome slow kinetics [5]. Similar to photoelectrochemical (PEC) water-splitting, we can suppose that there should be photo-bioelectrochemical water-splitting based on microalgal photoanode, in which absorption of visible light provides driven energy for water-splitting with the assistance of an applied bias.

In our lab, we developed a novel method to prepare microalgal photoanode, which can be applied to eukaryotic and prokaryotic alga, and investigated photoinduced electron transfer and photosynthetic oxygen evolution based on microalgal photoanode [6,7]. More than 60% of the electrons coming from water-oxidation in PS II could be extracted to generate photocurrent.

In this study, a microalga *Tetraselmis subcordiformis* (synonym: *Platymonas subcordiformis*)-based photoanode was prepared. By investigating effects of anode potential, cell density and BQ concentration on the photocurrent of microalgal photoanode, the preparation process of microalgal photoanode was optimized. In order to investigate the whole photo-bioelectrochemical process, dissolved oxygen near anode and dissolved hydrogen near cathode was measured by O_2 and H_2 microsensors respectively during electrochemical investigation of microalgal photoanode.

2. Materials and methods

2.1. Green microalga

T. subcordiform, a marine green microalga, was kindly presented by the Institute of Aquaculture of Liaoning Province, Dalian, China. The cells were grown in the airlift tubular photobioreactor bubbling with N_2 containing 2% (v/v) CO_2 at 27 °C in the following growth medium: 0.5 g KNO_3, 0.05 g KH_2PO_4, 0.81 g Tris, 0.33 mL glacial acetic acid, 1 mL of modified Walne medium, 1000 mL seawater from Yellow Sea in Dalian. Modified Walne Medium contained 0.8 g $FeCl_3$, 0.4 g $MnCl_2{\cdot}4H_2O$, 33.6 g H_3BO_3, 45.0 g EDTA·2Na, 20.0 g $NaH_2PO_4{\cdot}2H_2O$, 100.0 g $NaNO_3$, 0.021 g $ZnCl_2$, 0.02 g $CoCl_2{\cdot}6H_2O$, 0.009 g $(NH_4)_6Mo_7O_{24}{\cdot}4H_2O$, 0.002 g $CuSO_4{\cdot}5H_2O$, 1000 mL of water (Synergy water purification system, Millipore) [8]. The photon flux density focused on the airlift tubular photobioreactor was fixed at 200 μmol s^{-1} m^{-2} unless otherwise noted.

The chlorophyll content was estimated spectrophotometrically. The photosynthetic capacity of algal cell was measured by a chlorophyll fluorometer (Water-PAM WALZ, Germany) with the pulse-amplitude-modulation (PAM) [9].

2.2. Chemicals and instruments

All chemicals used in this study were of analytical reagent grade quality. Toray carbon fiber paper (TGP-H-060) was purchased from Toray Industries Inc. P-benzoquinone (BQ) was used as the exogenous artificial electron acceptor.

All electrochemical measurements in a single compartment cell containing a three-electrode configuration were carried out using electrochemical workstation (CS300, Wuhan Corrtest Instrument Co., Ltd, China). Microalgal photoanode was used as the working electrode and platinum electrode (2 mm × 7 mm) and Ag|AgCl|sat. KCl electrode was used as the counter and reference electrode, respectively. The electrochemical cell system was placed in an illumination incubator, and the fluorescent lamp was used as light source. Illumination by visible light was added from all directions of the cell. The light intensity focused on the fixed position of working electrode was fixed at 145 μmol s^{-1} m^{-2} unless otherwise noted. Electrolysis solution was 100 mL of sea water. The solution was deaerated by passing pure nitrogen gas before and during measurements. All measurements were carried out at room temperature.

2.3. Preparation of microalgal electrode

The porous silica sol was obtained by the hydrolysis followed by the condensation of tetraethoxysilane (TEOS) [10,11]. Briefly, silica sol was prepared as follows: 10 mL of TEOS, 60 mL of H_2O and 30 mL of 0.01 M HCl were violently stirred for 48 h at ambient temperature resulting in an acidic nanosol. Then the pH value of silica sol was adjusted to 7.5 by adding 1 M of NaOH. The silica sol was violently stirred for 48 h continuously.

The microalgal electrode was prepared as follows: cells were harvested by centrifugation at the speed of 500 g for 2 min. The algal cells pellet was uniformly dispersed in the 2 mL of silica sol. Then, 50 μL of resulting mixture was well coated on the paper carbon (1.2 cm × 2.0 cm) forming the thin layer, which was subsequently air-dried for 2 min.

2.4. Measurements of dissolved oxygen concentration and dissolved hydrogen concentration

The concentration of dissolved oxygen near anode and dissolved hydrogen near cathode was measured by O_2 and H_2 microsensors connected to a picoammeter (UniSense A/S, Aarhus, Denmark) respectively while electrochemical investigation of microalgal photoanode was carried out.

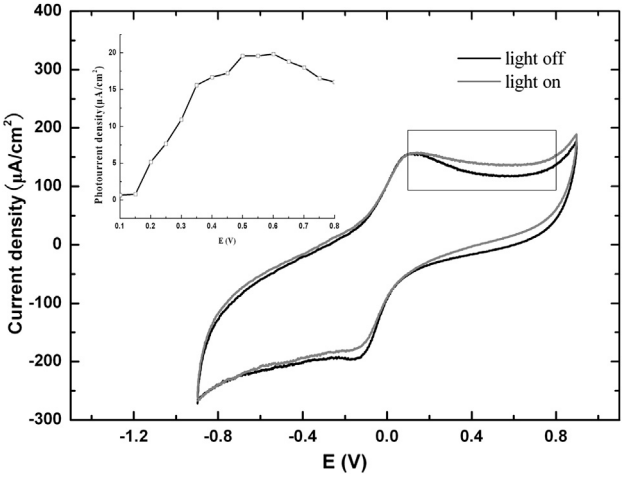

Fig. 1 – Cyclic voltammogram of microalgal photoelectrode without and with illumination. The scan rate was 50 mV s^{-1} with solution deaerated by nitrogen.

3. Results and discussion

3.1. The selection of the anode potential

Fig. 1 showed cyclic voltammogram curves of microalgal photoelectrode without and with illumination at a scan rate of 50 mV s^{-1} with solution deaerated by nitrogen and current increment curve from 0.1 V to 0.8 V.

As shown in Fig. 1, under illumination, the cathodic wave decreased and the anodic wave increased compared with that in dark condition. In the negative potential region, the oxidized form of BQ was reduced by electrons from both electrode and photosynthetic chain of algal cells under illumination, so there was a decrease in reduction current. In the positive potential region, the reduced BQ was oxidized at the electrode, but then the oxidized form was reduced by microalga, which caused an increase in oxidation current. From Fig. 1, it was observed that the photocurrent increased as anode potential increased from 0.15 V vs Ag|AgCl|sat. KCl and reached maximum value of between 0.5 V and 0.6 V vs Ag|AgCl|sat. KCl, and then decreased. So the anode potential was determined at 0.5 V vs Ag|AgCl|sat. KCl in the following experiments.

3.2. Effect of cell density on photocurrent

Various amount of algal cells were harvested by centrifugation and uniformly dispersed in the 2 mL of silica sol, then, 50 μL of resulting mixture was well coated on the paper carbon (1.2 cm × 2.0 cm) as microalgal electrode. The effect of immobilized cell density on photocurrent was shown in Fig. 2.

It was observed in Fig. 2 that photocurrent increased as the cell density increased at first and reached maximal value at 2.08×10^6 cells/cm^2, and then decreased. When the cell density was 6.24×10^6 cells/cm^2, the photocurrent decreased by 37.7% compared to the case of 2.08×10^6 cells/cm^2. Maybe the increase of cell density enhanced oxygen evolution from microalga, which caused that O_2 could not be displaced by N_2 in time when the cell density was more than 2.08×10^6 cells/cm^2. The electrons that BQ extracted from the photosynthetic electron transfer chain would be accepted by O_2 instead of working electrode. Similar result was also mentioned in Yagishita's work [12]. In addition, because the process was diffusion-limited, there would be a negative effect on photocurrent with further increase of cell density more than 2.08×10^6 cells/cm^2. Therefore, the optimum cell density immobilized on the working electrode was determined to be 2.08×10^6 cells/cm^2.

3.3. Effect of BQ concentration on photocurrent

The effect of BQ concentration on photocurrent was shown in Fig. 3. Photocurrent increased promptly from 6.73 μA/cm^2 at 20 μM to 34.37 μA/cm^2 at 200 μM with the concentration of BQ increasing, and then increased slowly between 200 μM and 300 μM. The photocurrent reached a maximum value of 37.0 μA/cm^2 at 300 μM, and then decreased to 30.48 μA/cm^2 at 500 μM. Because BQ competed with electron carrier of microalga for binding photosynthetic electrons, the improvement of BQ concentration would obviously enhance extraction of photosynthetic electrons from microalga, which means the improvement of photocurrent. The decrease of photocurrent may be because some damages happened to the photosynthetic system of algal cells when BQ was more than 300 μM.

By optimizing the preparation process of microalgal photoanode, the optimal photocurrent density of microalgae photoanode, 37.0 μA/cm^2, was achieved under illumination of 145 μmol s^{-1} m^{-2} at anode potential of 0.5 V vs Ag|AgCl|sat. KCl, immobilized cell density of 2.08×10^6/cm^2 and BQ concentration of 300 μmol/L.

3.4. Demonstration of photo-bioelectrochemical water-splitting

Fig. 4 showed the curves of oxygen evolution near anode, hydrogen evolution near cathode, current density and light intensity versus time.

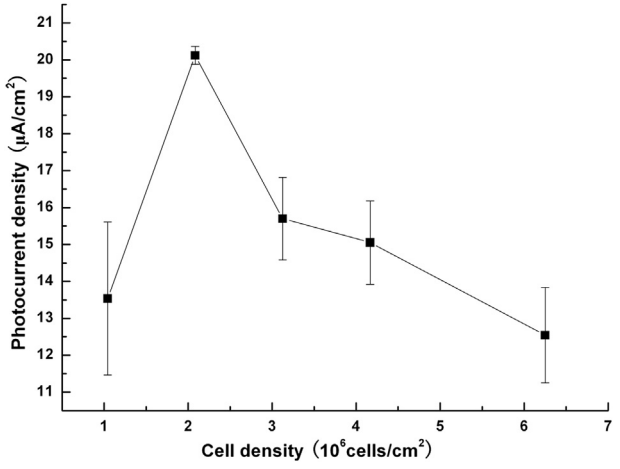

Fig. 2 – Effect of cell density on photocurrent. BQ concentration was 200 μM and the anode potential was 0.5 V vs Ag|AgCl|sat. KCl.

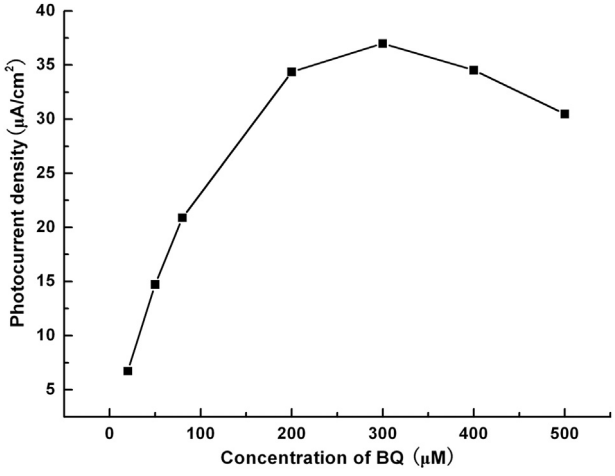

Fig. 3 – Effect of BQ concentration on photocurrent. Anode potential was 0.5 V vs Ag|AgCl|sat. KCl and cell density was 2.08×10^6/cm^2.

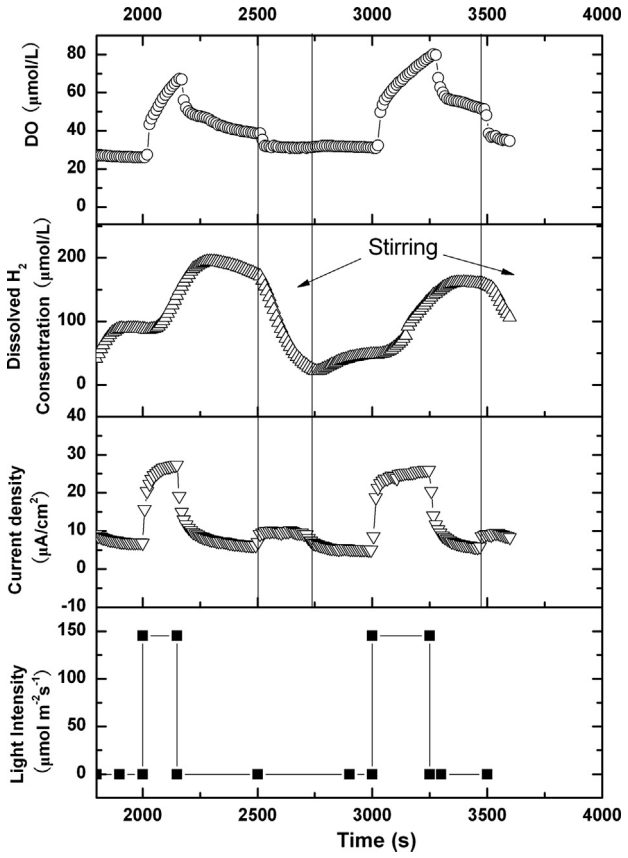

Fig. 4 – Curves of oxygen evolution at anode, hydrogen evolution at cathode, current density and light intensity versus time. Two illuminations were added at 2000 s and 3000 s and their lasting time were 150 s and 250 s, respectively. In order to recovering microsensors for next measurement, magnetic stirring was added between 2480 s and 2700 s and after 3450 s, respectively.

From Fig. 4, it was observed that oxygen concentration and current density always increased promptly with illuminations beginning, which means that oxygen evolution and photocurrent were synchronous to illumination. The increase values of oxygen concentration and current density were about 44 μM and 20 μA/cm^2 respectively. Compared with the cases of oxygen concentration and current density, hydrogen evolution process had some complex. In two measurements, hydrogen concentration had slow increases of about 46 μM and 27 μM within 500 s without illumination respectively. Under illuminations, there was an increase of about 1.6 μM in hydrogen concentration within initial 60 s, and then the hydrogen concentration rose promptly. During illuminations, the maximal increments of hydrogen concentrations were 107.5 μM and 113.2 μM respectively. The photo-hydrogen evolution was twice more than hydrogen evolution in dark condition. The increment of photo-hydrogen concentration was also twice more than that of oxygen concentration, which can be attributed to that cathodic area is five times less than anodic area. Obviously, the electrons for hydrogen evolution had two sources, microalgal metabolic process in dark condition and photosynthetic water oxidation, which resulted in the complexity of hydrogen evolution. The time lag of about 60 s between oxygen evolution and photo-hydrogen evolution can be due to the hydrogen diffusion between surface of cathode and H_2 microsensor. Ignoring the lag effect, photo-hydrogen evolution will also be synchronous to light impulse. The results demonstrated that a process of photo-bioelectrochemical water-splitting occurred in the three-electrode system based on microalgal photoanode. Hydrogen can be produced by the method.

4. Conclusion

Microalga *T. subcordiformis* (synonym: *Platymonas subcordiformis*)-based photoanodes were prepared by a novel method. By optimizing the preparation process, the optimal photocurrent density of microalgae photoanode, 37.0 μA/cm^2, was achieved under illumination of 145 μmol s^{-1} m^{-2} at anode potential of 0.5 V vs Ag|AgCl|sat. KCl, immobilized cell density of 2.08 × 10^6/cm^2 and BQ concentration of 300 μmol/L.

In a three-electrode system, the whole photo-bioelectrochemical process was investigated by electrochemical measurement and records of dissolved oxygen at anode and dissolved hydrogen at cathode. The electrons for hydrogen evolution had two sources, microalgal metabolic process in dark condition and photosynthetic water oxidation. The photo-hydrogen evolution was twice more than hydrogen evolution in dark condition. The oxygen evolution peak, hydrogen evolution peak and photocurrent response were all synchronous to light impulse in the three-electrode system. It demonstrated that a process of photo-bioelectrochemical water-splitting occurred in the three-electrode system based on microalgal photoanode. Hydrogen can be produced by the method.

Acknowledgments

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