

# The Characteristics of Dry *Purple Photosynthetic* Biofilm Used in Microbial Fuel Cells

Kaname Iwai, Dang-Trang Nguyen, and Kozo Taguchi

**Abstract**—Microbial fuel cells (MFCs) are a bioenergy source that can convert chemical energy into electrical energy using microorganisms. The inconvenient startup process is one of the main disadvantages of MFC technology. To improve this issue, the usability of dry purple photosynthetic bacteria (PPB) biofilm as the biocatalyst in an MFC was investigated in this study. This MFC composed of three parts: a paper-based membrane, an anode electrode with dry PPB biofilm, and a cathode electrode coated potassium ferricyanide. The dry PPB biofilm on the anode can be activated by adding water and generating on-demand electricity from photosynthetic activity. We measured the discharging voltage generated by the anodes stored for up to 8 weeks. The maximum power density of the MFC was  $2.90 \mu\text{W}/\text{cm}^2$  obtained at  $24.1 \mu\text{A}/\text{cm}^2$  current density.

**Index Terms**—Purple photosynthetic bacteria, dry biofilm, water-activated, long-term storage, potassium ferricyanide.

## I. INTRODUCTION

Currently, there is a great interest in using living microorganisms to produce electricity [1]. Compact, light-weight, and portable power source devices have attracted widespread attention in recent years due to the rapid increase in energy demand [2]. Miniaturizing MFCs are interesting technology for potentially powering portable electronic devices [3]. MFCs are emerging as green energy devices that can use bacterial metabolism for the generation of electrical current from a broad range of organic substrates [4], [5].

Photosynthesis is one of the important energy conversion pathway on earth, and it plays a key role in the biogeochemical cycles. In addition to plants, phototrophic microorganisms are regarded as the primary mediator for photosynthesis. Researchers have explored the reactions of photosynthetic process in the organisms for the past century. The mechanism will potentially contribute to the future new energy development [6]. During photosynthesis in MFCs, the microorganism captures solar energy in order to convert carbon dioxide and water into oxygen and carbohydrates. This generates organic matter which is then finally used in their respiration process by regenerating carbon dioxide and water. At this time, electrons are generated through the electron transport gateways and then they are discharged through the external electrical circuit. Released protons are then transferred from the anode to the cathode through a membrane, they re-associate with the electrons and oxygen to

reform water at the cathode. This process features as the essential ecosystem on the earth because they require only sunlight, water, and carbon dioxide to generate electricity which is considered to be a superior trait when compared to the other feasible sources such as photovoltaic [7].

Purple non-sulfur bacteria (PNSB) are a very versatile group of photosynthetic prokaryote microorganisms. These bacteria are capable of catalyzing hydrogen production under anaerobic conditions using nitrogenase with no need to protect their photosynthetic apparatus from inactivation from oxygen. In addition, they are capable of fixing atmospheric nitrogen via nitrogenase activity in the process. These PNSB are easily isolated from soil and water and they can grow as aerobes if oxygen is present to generate energy by respiration or as photoautotrophic organisms in the presence of light with the production of  $\text{H}_2$  by cyclic photophosphorylation [8]. Photosynthetic MFCs were originally tested in the 1960s with metal electrocatalysts and in the 1980s with artificial electron mediators in the anode chambers [9]. The device is a photosynthetic MFC, which can use phototrophic prokaryotes to convert light energy into electricity through photosynthesis process. The photosynthetic MFC is considered as the most promising energy-developed equipment in the future, which not only treat wastewater but also produce bioelectricity [6].

Several research have attempted to build a cathode compartment containing algae and separate it from the anode compartment by a cation exchange membrane [10], [11]. Real industrial applications of MFCs have been severely hindered by many problems such as the inconvenient startup process, low power output, expensive materials, and complicated reactor configuration. To address these issues, microalgae have been tested as a biocatalyst in the cathode chamber, depicting its feasibility to provide oxygen. Furthermore, a photosynthetic biocathode was also reported to increase the power generated in a sediment MFC [12].

In recent years, carbon nanotube (CNT) and graphene have been used as electrodes in fuel cells to further improve the performance. These carbon-based materials are generally coated on a suitable substrates such as carbon paper, carbon cloth, graphite, glass etc. to function as electrodes. More recently, paper has emerged as an inexpensive, convenient, and flexible platform causing a paradigm shift in the development of next generation energy conversion and storage devices such as fuel cells, batteries, and supercapacitors [13]. Paper-based electrodes are conventionally fabricated by methods such as screen printing, inkjet printing, spin coating, and oxidative chemical vapour deposition.

In this work, we created a compact MFC, which used dry

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PPB biofilm for on-demand electricity generation and reducing the inconvenient startup process. The PPB biofilm was pre-inoculated on a CNT-coated carbon sheet substrate.

## II. MATERIALS AND METHODS

### A. Experimental Setup

The experimental setup is shown in Fig. 1. The voltage between the anode and cathode was measured with a data acquisition system (DAQ, National Instrument, USB-6211), and recorded every 30 s via a customized LabView interface on a computer (PC). An external resistor (R) connected between the anode and cathode to discharge the MFC. The current through this resistor is calculated using Ohm's law.

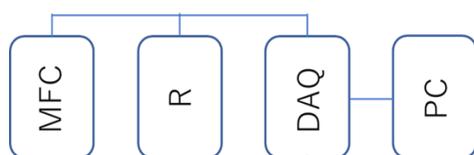


Fig. 1. Measurement setup.

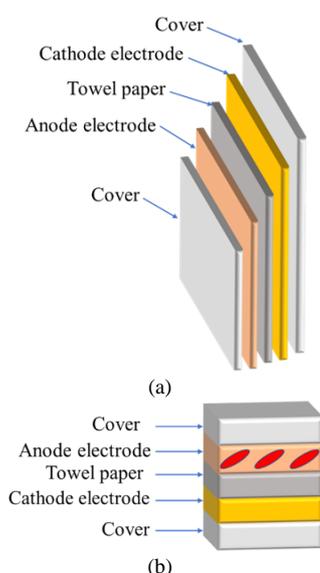


Fig. 2. The design of MFC: side view (a) and front view (b).

### B. MFC Design and Materials

The design of the MFC and its schematic diagram are shown in Fig. 2. We prepared the electrodes (10 mm × 10 mm) using hydrophilic carbon sheets (0.2 mm thickness, Azumi Filter Paper Co., Ltd). To make the anodes, the carbon sheets were dipped into CNT coating liquid (N7006L, KJ specialty Paper Co., Ltd.) for 1 min, and dried at 50 °C for 20 min. However, the as-fabricated electrodes became hydrophobic because of CNT. To make it hydrophilic, we conducted plasma treatment for 10h by an Ozone Killer (Filgen, UV253E) [14]. On the cathode side, we immersed the carbon sheets for 1min into the solution of 2 ml potassium ferricyanide 0.75 M and 20 ml CNT, which has been mixed for 24 h. Finally, the cathode was dried at 50 °C for 20 min. Paper-based proton exchange membrane (PEM) was made of towel paper (15 mm × 15 mm) (Monotaro, Japan), which

was coated by hydrophobic spray. The battery case was made of silicon rubber (15 mm × 15 mm) (2 mm thickness Monotaro, Japan). On the anode side silicon rubber was drilled a hole (5 mm × 5 mm) for dropping water to the anode.

### C. Biofilm Formation

PPB was inoculated in a special culture solution (Culture 1). Culture 1 contained PPB 1 ml, PSB medium 0.1 ml and purified water 8.9 ml. The culture conditions were at 30 °C under lamp light illumination for 2 weeks. Fig. 3 shows pictures of Culture 1 before and after incubation.

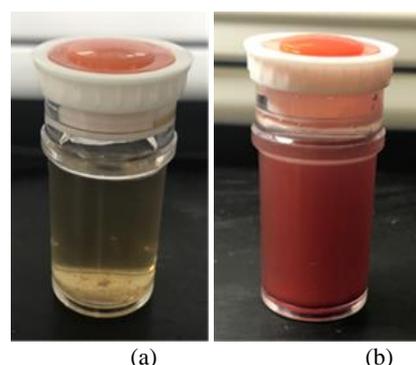


Fig. 3. The culture solution before incubation (a) and after incubation for two weeks (b).

From Fig. 3, it can be confirmed that Culture 1 turned red because of proliferating the number of PPB cells. The anode electrodes were immersed into Culture 1 for 4 days to make biofilm. As the next step, they were taken out of the culture solution and dried at room temperature for 48 hours (Fig. 4).

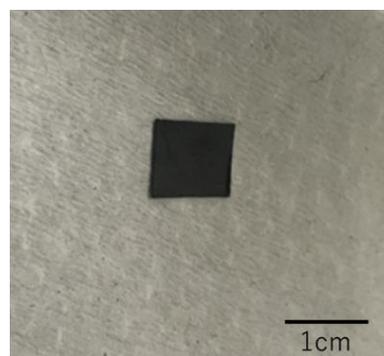


Fig. 4. Photo image of an anode electrode.

The dried biofilm anodes were stored in zip lock plastic bags and placed at room temperature indoor light. Surface morphology image of the anode electrodes with attached PPB biofilm was observed by scanning electron microscopy (SEM).

## III. RESULTS AND DISCUSSION

### A. SEM Images

The surface of the PPB biofilm formed on the anode electrode was observed as shown in Fig. 5. Fig. 5 (a) is the surface of carbon sheet coated CNT with plasma treatment (CS+CNT) and Fig. 5 (b) is the surface of carbon sheet (CS).

The size of bacterial cells is 2 ~ 3  $\mu\text{m}$ . From Fig. 5, it can be confirmed the increase of surface area of the CS-CNT anode compared with CS anode. The high surface area contributed to more adsorption of bacteria cells to the surface of the CS-CNT anode.

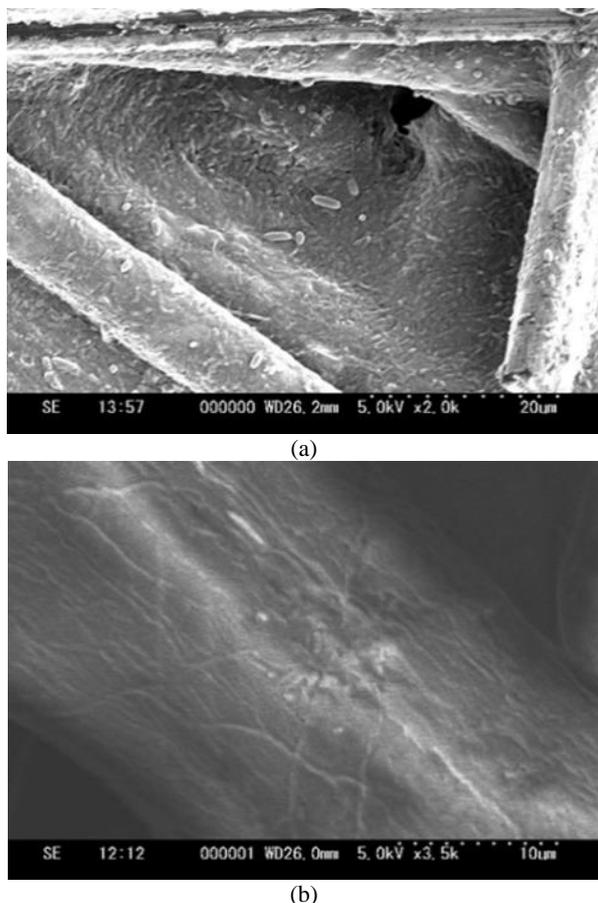


Fig. 5. SEM images of (a) CS+CNT biofilm anode and (b) CS biofilm anode.

### B. Power Density Measurement

In this paper, we focused on investigating the usability of dry PPB biofilm in MFCs, that could be activated by merely adding water and generating electricity from photosynthetic activity. The power density of the MFC was measured by varying various external resistors of 1 k $\Omega$ , 5 k $\Omega$ , 10 k $\Omega$ , and 50 k $\Omega$ . Fig. 6 (a) and (b) show the discharging voltage of the MFCs using CS+CNT and CS as the anode electrodes, respectively. It can be seen that the CS+CNT biofilm anode generated much higher the voltage than that of the CS biofilm anode. This result is attributed to the high surface area of the CS-CNT anode.

The power density was calculated based on the maximum discharging voltage obtained in Fig. 6. The calculated power density is shown in Fig. 7. It has been proved that the maximum power density of a device is obtained when the external resistance is equal to the internal resistance of the device [15]. Thus, the internal resistance of the MFC was about 5 k $\Omega$ . The maximum power density generated by the CS+CNT biofilm anode (2.90  $\mu\text{W}/\text{cm}^2$ ) was more than an order of magnitude higher than that of the CS biofilm anode (0.2  $\mu\text{W}/\text{cm}^2$ ). Based on this result, it can be confirmed that the performance of the MFC can be much improved by using the CS-CNT biofilm anode.

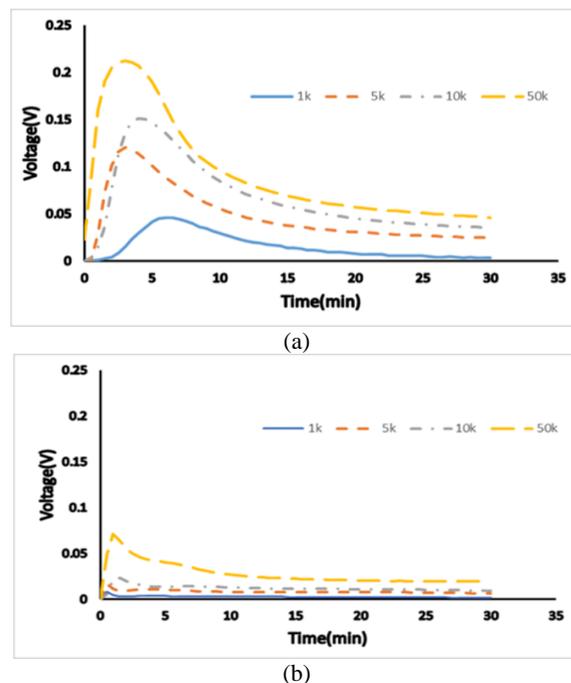


Fig. 6. The discharging voltage of the MFCs using the CS+CNT biofilm anode (a) and the CS biofilm anode (b).

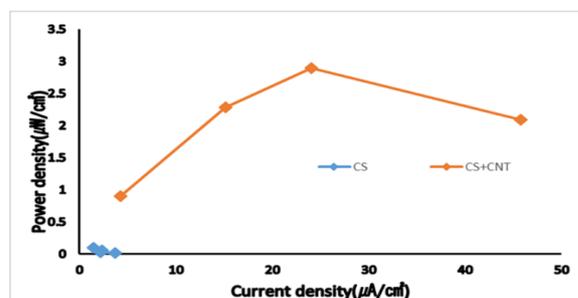


Fig. 7. The power density of the MFCs using the CS+CNT and CS biofilm anodes.

### C. Long Term Storage

By using the CS+CNT biofilm anode, we conducted an experiment to test the influence of long-term storage on the dry biofilm of PPB. The power density of the MFCs using the biofilm anodes stored for zero week (after drying), one week, two weeks, three weeks, and eight weeks were measured. The experimental result is shown in Fig. 8. The output decreased significantly after the first two weeks. However, the power density was quite stable with samples stored longer than two weeks. After storing for eight weeks, the anode could still generate significant power. In other words, PPB biofilm can still be alive and active after long-term storage in dry condition.

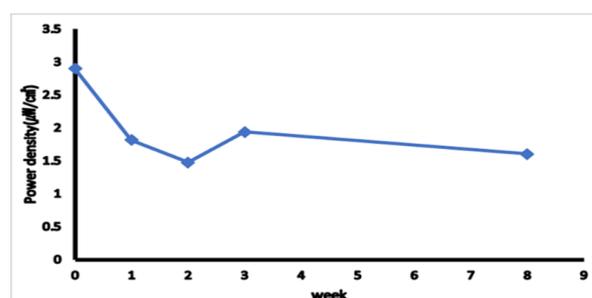


Fig. 8. The power density of the MFCs using the biofilm anodes as a function of stored time.

In this paper, we have succeeded in the demonstration of using the dry biofilm of PPB pre-inoculated on the anode of the MFC for instant and on-demand electricity generation. The biofilm anode can generate power even after stored for 8 weeks. In addition, the MFC is compact and made of low-cost materials. The method of using dry biofilm anode and activating by adding water presented here helps to reduce the inconvenient startup process in MFC applications.

#### IV. CONCLUSION

In this research, an MFC which used PPB as the biocatalyst in the anode was investigated. The biofilm of PPB pre-inoculated on the anode electrode demonstrated the advantage of long-term viable and active in dry condition. Experimental results showed that CNT-coated electrode could significantly improve the performance of the MFC. The dry PPB biofilm could generate electricity after eight weeks of storage. The maximum power density and current density of the MFC were  $2.90 \mu\text{W}/\text{cm}^2$  and  $24.1 \mu\text{A}/\text{cm}^2$ , respectively.

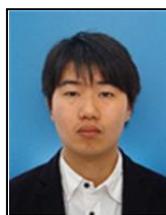
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