A flexible micro biofuel cell utilizing hydrogel containing ascorbic acid

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Abstract. This paper reports on a biofuel cell with a dimension of 13x24 mm² fabricated on a flexible polyimide substrate. Its porous carbon-coated platinum (Pt) electrodes of 3 mm in width and 10 mm in length were fabricated using photolithography and screen printing techniques. Porous carbon was deposited by screen printing of carbon black ink on the Pt electrode surfaces in order to increase the effective electrode surface area and to absorb more enzymes on the electrode surfaces. It utilizes a solidified ascorbic acid (AA) aqueous solution in an agarose hydrogel to increase the portability. The maximum power and power density for the biofuel cell with the fuel unit containing 100 mM AA were 0.063 μW and 0.21 μW/cm² at 0.019 V, respectively.

1. Introduction

Microelectromechanical Enzymatic biofuel cells that use glucose in a human body to produce electricity for embedded Microsystems have been of special interest. This is because they work under mild conditions of room temperature, neutral pH and atmospheric pressure, which make them amenable to the human body [1]-[14]. Nishizawa et al. reported a biofuel cell with a microchannel designed to continuously deliver fuel to the biofuel cell [15]. Fukushi et al. also reported a flexible glucose biofuel cell with a microchannel fabricated on a polyimide (PI) substrate [16]. However, these devices sometimes require complex biofuel delivering systems, which causes portability loss of the biofuel cells. We reported a portable simple-structured biofuel cell with planar two electrodes on a PI substrate that utilized agarose hydrogel containing glucose as a fuel [17].

Ascorbic acid (AA) also known as vitamin C, is an environmentally and biologically friendly compound. Thus, it is interesting to utilize an aqueous AA fuel absorbed in hydrogel. Typical hydrogels include collagen, polyacrylamide and agarose as polymers, and they are expected to store a AA aqueous solution. Collagen has been reported to be easily decomposed by ultraviolet rays in the sunlight [18], and it is not suitable for a portable hydrogel solid fuel. On the other hand, agarose has been reported to be harmless to human body, and the diffusions of AA and oxygen in agarose gels are almost the same as those in water [19]. Thus, we considered that agarose is suitable for a portable hydrogel solid fuel containing AA. In this paper, a AA biofuel cell with carbon electrodes modified with enzymes that utilizes an agarose hydrogel containing an aqueous solution of AA is reported.
2. Experimental

2.1 Design of Ascorbic acid fuel cells

Figure 1 shows the schematic view of the fabricated AA biofuel cell. The anode with an area of 3x10 mm² is covered by carbon, the cathode with an area of 3x10 mm² is covered by carbon modified by bilirubin oxidase (BOD), and the reference electrode with an area of 3x7 mm² is covered by Ag/AgCl. The solid agarose gel as a portable fuel containing 100 mM AA solution was placed on these electrodes. The size of the agarose gels was 13x 20 mm² with the thickness ranging from 1 to 4 mm.

Figure 2 shows the power generation mechanism. On the anode, the AA is oxidized at the anode electrode surface and dehydro ascorbic acid is generated. As a result, electrons and protons were produced on the anode. The electrons were transferred from the anode to the cathode via an external circuit generating electricity. Oxygen (O₂) in the fuel solution is reduced by BOD immobilized on the cathode to water on reaction with electrons and protons.

2.2 Ascorbic acid fuel fabrication process

A mixture of ascorbic acid, agarose (Agarose Basic, TAKARA), and a phosphate buffer solution (PBS) (50 mM, pH 7.0) was boiled to dissolve at 100 °C. The weight percentage (wt%) of agarose was varied for 0.5, 1.5, and 2.5 wt% to know the optimum condition for the agarose concentration.

2.3 Fabrication of enzyme-modified electrodes

The Pt wiring patterns were deposited on a 50 μm thick PI film (Kapton) using a mask in a radio-frequency sputtering equipment. The carbon-modified anode and cathode electrodes were patterned on the surfaces of the Pt film by screen printing using a carbon paste (MRX-713J-A, TAMURA) that was then cured at 150 °C for 15 min. Finally, the surfaces of the carbon paste film were coated by screen printing with Ketjenblack EC 300J (KB, Lion Inc.), and polyvinylidene fluoride (PVDF, KUREHA Inc) mixed ink was then cured at 80 °C for 20 min. Ag/AgCl reference electrodes were patterned on the surfaces of Pt film by screen printing with Ag/AgCl ink (BAS Inc.) that was then cured at 120 °C for 5min. The increased effective specific surface area increases the amount of enzyme adsorbed to the electrode surface was modified electrode surfaces in KB. The preparations of enzyme coated electrodes are described as follows. BOD with an amount of 1 mg was mixed with a 25 μl PBS (50 mM, pH 7.0). Then, a 2 μl portion of the resulting solutions was put onto the cathode, and left to dry for 20 min.

Figure 3 shows photographs of (a) a fabricated biofuel cell with a solidified AA aqueous solution on the electrodes and (b) an agarose AA fuel with agarose weight ratio 2.5 wt%. Exudation of water was not observed when holding the fuel with fingers, which suggests the portability of the fuel.
measurements

Cyclic voltammogram (CV) measurements were performed with the agarose hydrogel containing AA 100 mM on the anode and the cathode at room temperature. As a reference, these electrodes were immersed in an aqueous solution containing 100 mM AA. When the CVs of the anode were measured, the surface of the cathode Pt area was not covered by carbon and enzymes, and the Pt area was used for the counter electrode. When the CVs of the cathode were measured, the surface of the anode Pt area was not covered by carbon and enzymes. The reference electrode was prepared by coating the reference electrode area with the Ag/AgCl ink. The electrochemical reactions within the biofuel were investigated by plotting the oxidation or reduction current densities \( J \) of the anode and the cathode. The oxidation and reduction current densities were analyzed by the Randles-Sevcik equation [20] below;

\[
J = 2.99 \times 10^5 n(\alpha n)^{1/2}AD^{1/2}C^{1/2}v^{1/2}
\]  

(1)

where \( \alpha \) is transfer coefficient, \( n \) is the number of reaction electrons, \( A \) is the electrode area, \( D \) is the diffusion coefficient of fuel or oxygen, \( C \) is the concentration of fuel or oxygen, \( v \) is the potential sweep rate.

The generated power was evaluated by measuring the cell voltage while varying the external load resistance between 0 and 2 MΩ. The power was derived using the following equation;

\[
W = \frac{V^2}{R}
\]  

(2)

where \( R \) is the load resistance, and \( V \) is the generated voltage appearing between the terminals of the fuel cell (see Fig.1). The power density was calculated by dividing the 30 mm\(^2\) of the anode area.

3. Results and discussion

Figure 4 shows the CV curves of the anodes using the solidified hydrogel containing 100 mM AA and agarose with different concentrations from 0.5 wt% to 2.5 wt%. The CV curve for the anode in the AA PBS solution in 50 ml of air-saturated 100 mM AA solution is also shown for comparison in Fig. 4. These measurements were done at the potential sweep rate of 20 mV/s. The CV curves for these 4 devices are very similar. Thus, it can be said that the oxidation of AA near the anode is very similar, and the agarose concentration has minimal impact on AA oxidation. Figure 5 shows the CV curves of the cathodes using the solidified hydrogel containing 100 mM AA and agarose with different concentrations from 0.5 wt% to 2.5 wt%. The CV curve measured in 50 ml of air-saturated 100 mM AA solution is also shown in Fig. 5. These measurements were done at the potential sweep rate of 20 mV/s.

The reduction peak current densities at -0.8 V for the biofuel cells with the agarose hydrogels were very similar. The agarose concentration has minimal impact on reduction of oxygen. However, the reduction current density of the cathode for the biofuel cell immersed in the 100 mM AA solution in a beaker showed much larger reduction current due to the larger amount of dissolved oxygen.
Figure 4. Cyclic voltammograms for the anode in 100 mM AA PBS and the anodes contacted with agarose gel fuel containing 100 mM AA with agarose concentration 0.5 wt%, 1.5 wt%, and 2.5 wt%.

Figure 5. Cyclic voltammogram for cathode in 100 mM AA solution and agarose gel of agarose concentration 0.5 wt%, 1.5 wt%, and 2.5 wt%.

Figure 6 shows the relationships between the generated current density and the voltage across the load resistor for the biofuel cells with the biofuel cell units containing agarose with different concentrations. They were also compared with the biofuel cell in a 100 mM AA PBS in a 50 ml beaker. The generated currents suddenly dropped when the voltage increased, and stayed above 0.5 V. There was almost no difference within these biofuel cells in the early stage of the biofuel cell operation. It should be noted that the current density of the fuel cells during the power generation is different from those of the CV curves where the voltages are scanned from outside.

Figure 7 shows the relationships between the power density and the voltage. The maximum power and power density for the biofuel cell with the fuel unit containing 2.5 wt% agarose were 0.063 μW and 0.21 μW/cm² at 0.019 V, respectively. There are almost no differences on the power generation performances within the biofuel cells investigated here. Therefore, the optimal concentration of agarose was determined to be 2.5 wt% in order to keep the shape of the AA fuel units, which increased the portability of the fuel units.
4. Conclusion
A portable biofuel cell with a cathode modified with bilirubin oxidase was fabricated on a flexible polyimide substrate. The anode and the cathode with an area of 3x10 mm$^2$ were separated with a gap of 1 mm. The portable AA biofuel was prepared by solidifying a 50-200 mM AA solution containing agarose. The maximum power and power density were 0.063 μW and 0.21 μW/cm$^2$ at 0.019 V, respectively when the solidified hydrogel biofuel with a dimension of 13x24x4 mm$^3$ containing a 100 mM AA solution and 2.5 wt% agarose was used.

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References
[1] N. Yuhashi, M. Tomiyama, J. Okuda, S. Igarashi, K. Ikebukuro and K. Sode. 2005 Biosens. Bioelectron. 20 2145
[2] I. Willner and E. Katz. 2005 Bioelectronics, Wiley-VCH, Weinheim.
[3] G.T.R. Palmore, H. Bertschy, S.H. Bergens and G.M. Whitesides. 1998 J. Electroanal. Chem. 443 155
[4] A. T. Yahiro, S. M. Lee, D. O. Kimble. 1964 Biochim. Biophys. 88 375
[5] K. C. Swades and R. L. Derek. 2003 Nature Biotechnology: 21 1229
[6] D. R. Bond and D. R. Lovley. 2003 Appl. Environ. Microbiol. 69 1548
[7] M. J. Moehlenbrock and S. D. Minteer. 2008 Chem. Soc. Rev. 37 1188
[8] K. G. Lim and G. T. R. Palmore. 2007 Biosens. Bioelectron. 22 941
[9] Y. Kamitaka, S. Tsujimura, N. Setoyama, T. Kajino and K. Kano. 2007 PCCP Phys. Chem. Chem. Phys. 9 1793
[10] S. Tsujimura, K. Kano and T. Ikeda. 2002 Electrochemistry. 70 940
[11] S. Tsujimura, M. Fujita, H. Tatsumi, K. Kano and T. Ikeda. 2001 PCCP Phys. Chem. Chem. Phys. 3 1331
[12] T. Chen, S. C. Barton, G. Binyamin, Z. Q. Gao, Y. C. Zhang, H. H. Kim and A. Heller. J. Am. Chem. Soc. 123 8630
[13] F. Sato, M. Togo, M. K. Islam, T. Matsue, J. Kosuge, N. Fukasaku, S. Kurosawa and M. Nishizawa. 2005 Electrochem. Commun. 7 643
[14] S. C. Barton, J. Gallaway and P. Atanassov. 2004 Chem. Rev. 104 4867
[15] M. Nishizawa, M. Togo, A. Takamura and T. Abe. 2005 Proceedings of the fifth international workshop on micro and nanotechnology for power generation and energy conversion applications
[16] Y. Fukushi, S. Koide, R. Ikoma, W. Akatsuka, S. Tsujimura and Y. Nishioka. 2013 J. Photo. Polym. Sci. Tech. 26 303
[17] H. Goto, Y. Fukushi, and Y. Nishioka. to be published in IEICE Trans. Electtronics.
[18] S. Sakura, D. Fujimoto, K. Sakamoto, A. Mizuno and K. Motegi. 1982 Can. J. Biochemistry. 60 525
[19] A. Polson and D. G. Parkyn. 1969 Biopolymers. 7 107
[20] A.J. Bard and L.R. Falkner. 2001 Hoboken: Wiley and Sons. 226