Polypyrrole-Coated Reticulated Vitreous Carbon as Anode in Microbial Fuel Cell for Higher Energy Output

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A microbial fuel cell is a noble 'green' technology generating electricity from biomass and is expected to find applications in a real world. One of main hurdles to this purpose is the low power density. In this study, we constructed a prototype microbial fuel cell using *Proteus vulgaris* to study the effect of various reaction conditions on the performance. Main focus has been made on the modification of the anode with electropolymerized polypyrrole (Ppy). A dramatic power enhancement was resulted from the Ppy deposition onto the reticulated vitreous carbon (RVC) electrode. Our obtained maximum power density of 1.2 mW cm^{-3} is the highest value among the reported ones for the similar system. Further power enhancement was possible by increasing the ionic strength of the solution to decrease internal resistance of the cell. Other variables such as the deposition time, kinds of mediators, and amount of bacteria have also been examined.

Key Words : Microbial fuel cell, Polypyrrole modification, Ionic strength, Power density

Introduction

A Microbial fuel cell (MFC) is a device that converts chemical energy to electrical energy using bacteria as bio-catalysts.[1-5] Bacteria at an anodic compartment oxidize variety of organic matters such as carbohydrates or acetate to produce electrons. Thus produced electrons are transferred to the cathode through the external load to reduce electron acceptors in a cathodic compartment to produce electricity. Since MFC can use renewable biomasses, it has been known as environmentally friendly and sustainable. As a 'green' technology, renewed interest has recently been poured upon MFCs that utilize not only conventional heterotrophic cells but also photoheterotrophic cells[6] and even sediment cells.[7] Depending on the operating types, there have been developed several types of MFCs which include mediator type fuel cells,[8] mediator-less fuel cells,[9] and membrane-less fuel cells.[10] Although MFCs still fall short of alternative energy source, they can find applications in wastewater treatment[11] and power supply for remote sensors using indigenous fuels.[12]

The main hurdle to commercialization of MFCs as a power source is their low power output although coulombic efficiency easily reaches over 80%.[13] Contrary to the other fuel cells such as inorganic fuel cells or enzyme fuel cells, where the chemical reactions take place at high rate, the slow reaction rate is the intrinsic problem of a MFC since substrates undergo long metabolic pathways to be utilized. Despite this fact, general consensus is that there is still much room for power output enhancement. A lot of works have been done for this purpose, which include acclimation of the inoculum,[14] reduction of the electrode spacing,[15] increasing solution conductivity by varying the solution ionic strength,[16] tuning the initial culture conditions,[17,18] and using new cathodic electron acceptors.[19] At the same time, many researchers have focused on improving electrode materials. Focus has been made both on cathodic and anodic materials. Many new cathodic materials based on non-noble metal oxygen catalysis instead of platinum had been reported due to its high price.[20,21] Efforts made on the anode, on the other hand, were immobilizing a mediator,[22] treating the anode with ammonia,[23] and using a composite electrode as an anode.[24]

In this paper, we describe polypyrrole (Ppy) can be a good candidate for the power enhancement when it is used to coat reticulated vitreous carbon (RVC). Ppy, since it was first electrochemically synthesized more than two decades ago,[25] has attracted great interest in its potential applications in preparing actuators, chemical sensors, biosensors, electrodes, and electronic devices,[26-30] because of its environmental stability, ease of synthesis, and high conductivity at room temperature.[25] More recently, Ppy has found its applicability to remove negatively charged colloids,[31,32] such as clay particles and humic acid, after it was coated on high surface area carbon substrates like carbon fiber and RVC. RVC is an ideal material as an anode for MFCs in that it is chemically inert, very rigid, and has high surface area and good electric conductivity. Most importantly, it provides an open pore structure so that the bulk of RVC could be accessed by the reactants.[33]

Experimental

Preparation of microorganism. *Proteus vulgaris* (ATCC 6059) was obtained from KCTC (Korean Collection for Type Cultures) and maintained on a nutrient agar plate at 4 °C. *P. vulgaris* was aerobically grown in a nutrient badge containing 3 g L^{-1} of Yeast extract and 5 g L^{-1} tryptone at 37 °C. The cells were harvested by centrifuging at 8000 g for 10 min and washed with 0.1 M phosphate buffer solution (pH 7.0). The washed microorganisms were resuspended in the same phosphate buffer solution for the experiments. Glucose

was used as a fuel.

Fuel cell setup. The fuel cell used in our experiments was made of Plexiglas and composed of anodic and cathodic compartments (internal dimension of 10 mm × 10 mm × 5 mm) separated by the cation exchange membrane (Nafion 117, Aldrich). Ppy-coated RVC and bare RVC with 100 pores per inch (ppi) were used as an anode and a cathode, respectively. The anode RVC had a dimension of 0.7 mm × 0.7 mm × 0.2 mm. In our experiments, we employed a mediator-type MFC where thionin, methylene Blue, and neutral Red were used as a mediator. For a cathodic reaction, we used bare RVC and ferricyanide (0.1 M) as a cathode and an electron acceptor, respectively. RVC was washed with 75% alcohol and deionized water, dried at 70 ℃ in an oven, and then placed in a phosphate buffer solution (PBS, 50 mM) before use. Both anode and cathode compartment contained 0.1 M phosphate buffer solution (pH 7.0) as electrolyte.

Nafion 117 was successively pretreated by boiling it in 3% hydrogen peroxide for 1 h and in 1.0 M sulfuric acid for another 1 h in boiling deionized water, and then stored in deionized water prior to use.

The cell discharge was done by putting external load between anode and cathode. The cell potential was measured using an automatic battery cycler (WBCS 3000, WonAtech, Korea) for the different load as function of time. Since the anode is a three-dimensional electrode, we expressed our results per unit volume of the anode.

Electrodeposition of polypyrrole on RVC. Polypyrrole film was electrodeposited on RVC surface from an aqueous solution containing 0.1 M pyrrole and 0.1 M KCl by applying 0.9 V for 15 min. After washing it with deionized water, Ppy-coated RVC was dried at 70 °C in an oven. SEM images were taken before and after coating.

Results and Discussion

Effect of electrodeposition time on the performance of MFC. Figure 1 shows the SEM images of RVC before (a) and after (b) Ppy electrodeposition. RVC (100 ppi) has a three-dimensional network type structure with several hundred micron size pores interconnected by carbon fibers. These holes are, however, much too big for the bacteria to reside inside. Upon electropolymerization, carbon fibers begin to be covered by Ppy, making the surface rougher and pores smaller so that it offers an ideal environment for bacteria.

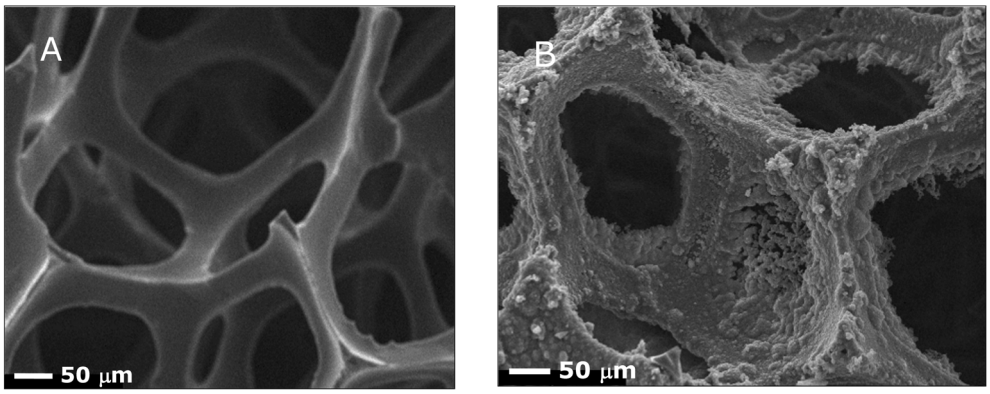


Figure 1. SEM images of RVC before (A) and after (B) electropolymerization of pyrrole for 15 min.

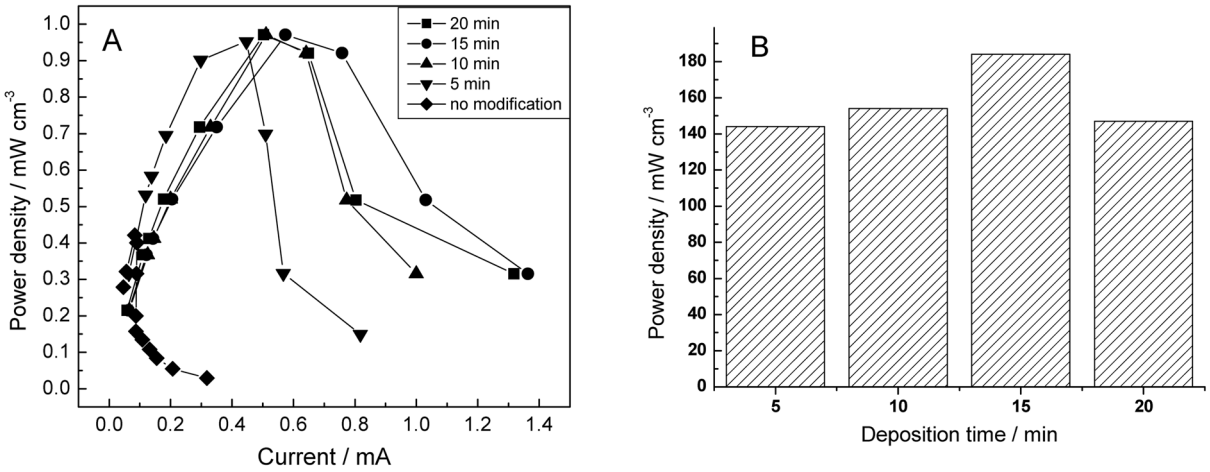
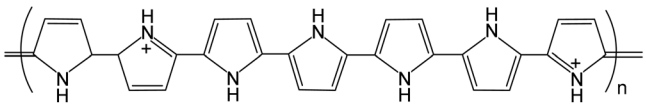


Figure 2. Polarization curves (A) and power density vs deposition time (B) for a MFC comprised of *P. vulgaris* (80 mg mL^{-1}), methylene Blue (0.5 mM), and glucose (0.01 M) for different deposition time of 0 (◆), 5 (▼), 10 (▲), 15 (●), and 20 min (■). The solutions were buffered at pH 7.0.

Extensive polymerization blocks the pores.

First we studied the effect of polymerization time on the cell performance (Fig. 2A). As deposition time increases, the overall power density goes higher. Without modification, the power density reaches the maximum value of only 0.42 mW cm^{-3} at 0.08 mA. With more current flowing, power density drops abruptly, making this fuel cell unrealistic for the application. But the performance was dramatically improved with Ppy coating. The power density linearly increased until 15 min deposition time. The maximum power of 1.2 mW cm^{-3} at 0.58 mA was obtained. Longer deposition time, however, did not improve but rather deteriorated the performance (Fig. 2B). The positive effect of Ppy coating on power generation could be explained as follows: First Ppy coating increases effective surface area of RVC so that more bacteria and mediator molecules can have access to the surface, delivering more electrons to the anode per unit time. Bacteria in the bulk electrolyte cannot contribute to the performance although they keep oxidizing substrates. More importantly, positively charged nature of Ppy increases adhesion of negatively charged bacteria to the surface through electrostatic attraction,[23] thus making facile electron transfer to the anode.



The similar strategy has been adapted by Logan when he treated carbon cloth anode with ammonia to impart positive charge on the surface.[32] He observed enhanced power generation. Another possible explanation is that Ppy could function as a mediator itself. The long Ppy chain can penetrate the bacterial cell membrane to intercept electrons from the metabolic pathway to the anode. Furukawa *et al.*[34] reported enhanced performance when they coated carbon fiber with polyaniline in direct photosynthetic/metabolic biofuel cell. Ppy-coated RVC electrodes were used to see the effect of other variables on the fuel cell performance.

Mediators and performance. Fuel cell performance is greatly enhanced by employing mediators in most MFCs except for MFCs that use *Shewanella putrefaciens* where cytochromes are localized to the outer membrane to make it possible to directly transfer electrons to the electrode.[35] Mediators are interacting with the metabolic pathway of the biocatalyst to shuttle the electrons from intracellular space to extracellular environment, *i.e.*, the anode. Among many different types of mediators we chose thionin and methylene Blue (MB) as phenothiazine derivatives, and neutral Red (NR) as a phenazine molecule. These molecules have been known functioning as good mediators. Figure 3 shows that the performance is in the order of methlylene Blue, thionin, and neutral Red. It is very interesting to see that this order is the same as the one reported by Ieropoulos *et al.*[36] in which they tested different types of mediators for a prototype MFC using *E. coli*. They ascribed this fact to the difference in internal resistance (R_{int}). Internal resistance is a pure loss factor in any electrochemical cells, degrading the overall cell performance by the equation, $E_{cell} = E_{cathode} - E_{anode} - IR_{int}$. MB gave the lowest internal resistance while NR showed the

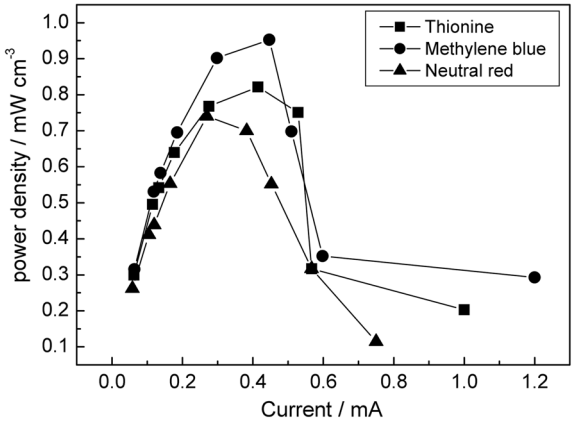


Figure 3. Polarization curves obtained when different mediators were used. Other conditions are the same as Fig. 2. Mediators (0.5 mM) are thionin (■), methylene Blue (●), and neutral Red (▲). Electrodeposition time was 5 min.

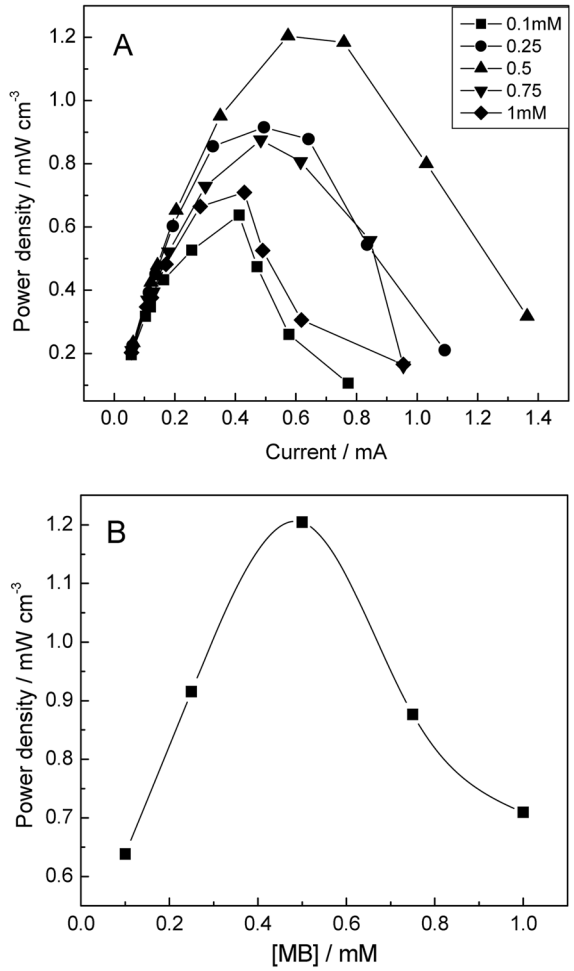


Figure 4. Polarization curves (A) and plot of power density vs methylene Blue concentration (B). MB concentration are 0.1 mM (■), 0.25 mM (●), 0.5 mM (▲), 0.75 mM (▼), and 1 mM (◆). Other conditions are the same as Fig. 2. Electrodeposition time was 15 min.

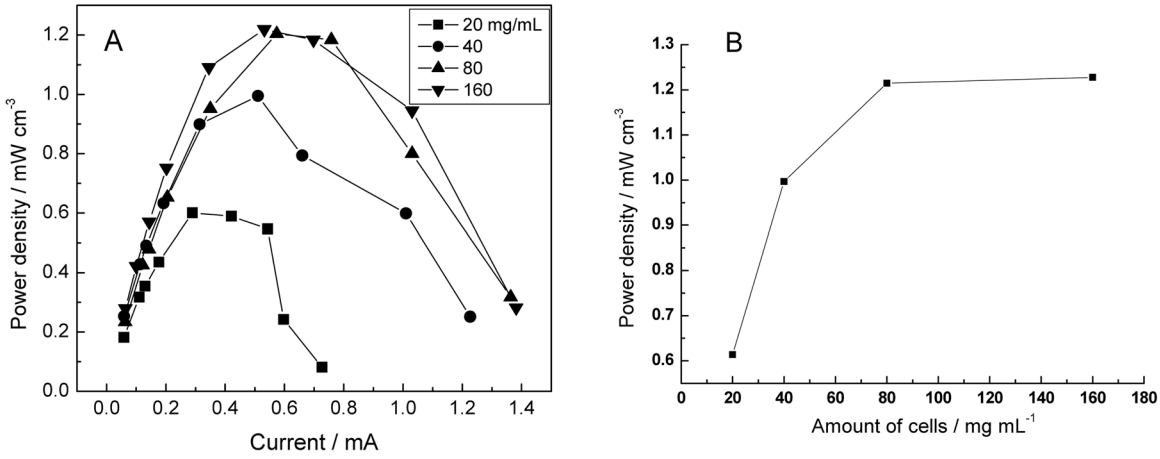


Figure 5. Polarization curves (A) obtained for different amount of bacterial cells: 20 mg mL^{-1} (■), 40 mg mL^{-1} (●), 80 mg mL^{-1} (▲), and 160 mg mL^{-1} (▼). Panel B was constructed from panel A. Other conditions are the same as Figure 2.

highest value. Another possible reason why NR is the worst mediator is that it most likely interacts with fermentation pathway,[37] not with respiration pathway. The higher efficiency is possible when mediators interact with the latter to accept more electrons.

The optimum mediator (MB) concentration was also investigated with all other conditions fixed (Fig. 4). Up to 0.5 mM, the power density increased with the concentration, reaching the maximum at 0.5 mM. This is due to the fact that the electron transfer reaction rate is proportional to the number of mediators. On the contrary to our expectation, however, at the higher concentrations than 0.5 mM, the power density decreased with the increase in concentration, indicating there is optimum mediator concentration. Too high concentration may have toxicity to the bacteria or the unreduced MB hinders the electron transfer from being adsorbed on the bacterial cell membrane.

Effect of the amount of bacteria. The number of bacteria was also shown to affect the power output when other conditions were fixed (Fig. 5). The power density increased rapidly with the increasing number of bacterial cells and then leveled off. In our case, 80 mg mL^{-1} was the minimum amount of *P. vulgaris* for the maximum power. 10 mg (dry weight) mL^{-1} solution contains *ca*. 2×10^8 cells. The fact of no further increase with the larger number of cells indicates that there is optimal effective number of bacterial cells for a given electrode geometry and fuel cell configuration. In this study we used 80 mg mL^{-1}.

Effect of ionic strength. Power density could be increased by reducing the internal resistance of the cell.[16] There are several ways to do this,[38] one of which is to decrease the distance between anode and cathode. Here we changed the ionic strength of the anolyte and catholyte by adding NaCl solutions of different concentrations. It gave rise to a dramatic difference (Fig. 6). When 100 and 300 mM NaCl was added, power density increased over 2.2 mW cm^{-3} from 1.2 mW cm^{-3} obtained for the MFC without NaCl. However, adding more NaCl rather resulted in power density decrease

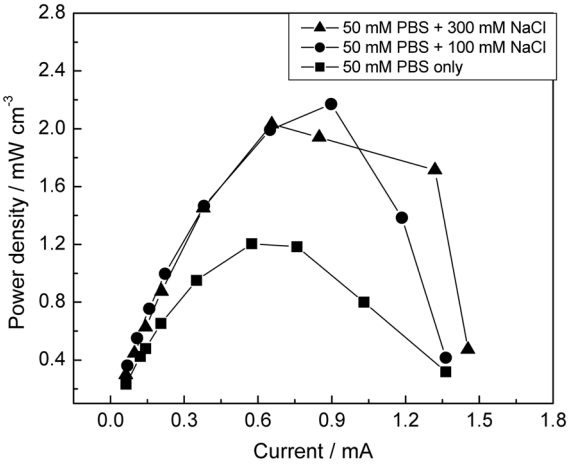


Figure 6. Polarization curves obtained for different ionic strength of NaCl: No NaCl (■), 100 mM (●), 300 mM (▲). *P.vulgaris*: 80 mg mL^{-1}. Phosphate buffer concentration was maintained at 50 mM. Other conditions are the same as Figure 2.

despite the reduced internal resistance. This is due to the simple fact that bacteria do not grow well in solutions of high salt concentration.

Our obtained maximum power density is the highest among those reported in literatures using *P. vulgaris*.[39,40] Using other reaction conditions or other cell configurations, power could be much enhanced. For example, Wilkinson *et al*. reported two mixed mediator system composed of MB and NR gave higher performance than when a single mediator (MB or NR) was used.[37] Schröder and coworkers[41] showed that use of a platinized carbon anode with polyaniline overlay boosted current output by more than one order of magnitude. Logan[15] and Liu[38] recently showed that by improving the cell configuration, the power density and coulombic efficiency could be increased by several factors. Adapting these progresses into our system, we hope to develop MFCs generating enough electricity for the practical applications.

Conclusions

A positively charged RVC surface was produced by polypyrrole coating. Higher power density was achieved on this electropolymerized RVC anode. The optimum operating reaction conditions have been examined, including the electrodoposition time of Ppy, the choice of mediators, the amount of bacterial cells, and the ionic strength. Our obtained maximum power density of 2.2 mW cm^{-3} was the highest value to our knowledge among similar systems. Further development for even higher power density is on the progress from a view point of fuel cell configuration and bacteria immobilization.

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