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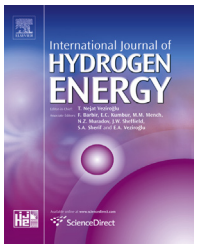

Effect of influential factors on microbial growth and the correlation between current generation and biomass in an air cathode microbial fuel cell

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ABSTRACT

Microbial biomass in a microbial fuel cell (MFC) was determined at varied external resistances and temperatures, and the current generations were correlated with biomass concentration. MFC operation with wastewater generated the highest current of 0.23 mA at 300 Ω compared with 100 Ω (0.22 mA) and 1000 Ω (0.18 mA). The microbial biomass (0.091 mg-protein/cm^2) on the anode with 100 Ω was higher than the other resistances. With growth media, current and biomass density were increased due to high strength electrolyte and nutrient. The MFCs with higher current generation contained a more amount of biomass on an anode electrode. The highest biomass growth rate with growth media was 1.307×10^{-4} mg-protein/hr·cm^2 at 30 °C (with 300 Ω), which was about 2 times higher than with wastewater. In linear regression, current generation is expected to increase by about 3 times as the biomass is doubled on the anode electrode.

Introduction

Wastewaters treatment is one of major practices in every day to day life for the human and environmental safety. In this regard, several inventions like anaerobic digestion, aerobic digestion and trickling filters were successfully operated. However, the sludge (bacterial biomass) generations from these technologies could increase the operational cost of entire wastewater treatment cost up to 25%–65% [1]. Microbial fuel cells (MFCs) are bio-electrochemical and innovative systems which can treat wastes and wastewaters with generation of simultaneously electricity [2]. In general, the organic compounds present in the wastewaters was consumed by electrogenic bacteria (exoelectrogens) present on the anode electrode with production of electrons and protons [3]. These produced electrons and protons then move towards cathode through external circuit and membrane where they get reduced to water [4]. Moreover, it was well known that MFC could generate less sludge generation compared to conventional treatment methods [1].

The sludge (bacterial biomass) formation in MFC is important parameter that should be considered while operating with different wastewater and operation conditions. In fact, most of the MFC studies were concerned to the generation of electricity with simultaneous COD and nutrient removals [5]. Up to now, only a few studies have been focused on

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the estimation of sludge generation in correlation with MFC voltage and bacterial biomass density on anode electrode [6]. However, these studies failed to measure the biomass growth on both anode and anolyte (planktonic) with respective to time and SCOD removals. Moreover, effect of external resistance has been reported previously [5,6], but there is no previous reports about temperature effect on biomass generations in MFC anode and anolyte with respective to time and SCOD removals.

In this study, effect of different external resistances (100 Ω, 300 Ω, and 1000 Ω) and temperatures (20 °C, 30 °C, and 35 °C) on microbial growth (anode electrode and anolyte) and power generation in single chambered air-cathode MFC were investigated. During this study, the microbial growth in duplicate MFC reactors anode electrode and anolyte were studied using both domestic wastewater and growth media (GM). The protein concentrations were measured with help of protein assay (BCA method) and used to calculate the biomass densities. The change in biomass growth of MFC (anode and electrode) was measured respective to time and SCOD removals. Finally, a correlation was made in between MFC voltage, Coulombic efficiency, biomass density on anode electrode, anolyte and SCOD removals with respective to applied external resistances and temperatures.

Materials and methods

Inoculation and media

Initially, the air cathode MFC anode was filled with 250 ml or 270 ml (based on experimental condition) of collected domestic wastewater from Giheung Respia wastewater treatment plant (Yongin-si, Korea). This wastewater was used as both inoculum and medium with 7 mM of anhydrous sodium acetate as an additional carbon source. Once a stable biofilm formation was observed and experimental analysis was completed with wastewaters from duplicate MFC reactors, the reactor's media was changed from wastewater to the GM media (growth media). The GM media was prepared with addition of vitamins, minerals into 100 mM phosphate buffer solution (5.883 g of KH_2PO_4 and 10.05 g of K_2HPO_4 in 1000 ml distilled water) as previously described [7] and 7 mM of sodium acetate anhydrous was used as carbon source.

Reactor configuration and operation

A single-chambered bottle type air cathode MFC, which are made of glass, were used in this study and all experiment runs were conducted with duplicate MFC reactors. The total MFC reactor volume was 280 ml, during testing effect of external resistance on biomass growth of anode 270 ml working volume was used. Whereas, while testing the effect of temperature on biomass growth of anode, 250 ml working volume was used. In all the experiments plain carbon paper having a total surface area of 48.16 cm^2 (8.6 cm × 2.8 cm) was used as anode electrode. Whereas, a 10% platinum coated carbon cloth (4.52 cm^2; Fuel Cell Earth, USA) was used as cathode electrode. Pretreated Nafion-117 was used as a separator in this study and the membrane pretreatment was carried out as previously described [7,8]. Copper wires were used to make electrical connections, and an external resistance of 1000 Ω was loaded in between anode and cathode. However, this applied external resistance was varied from 0 Ω (open circuit) to 100 Ω, 300 Ω and 1000 Ω during testing the effect of external resistance on MFC biomass growth. Initially, the MFC anode was fed with wastewater having 7 mM of sodium acetate, and then purged with pure nitrogen gas for approximately about 10 min to create anaerobic environment. Then the anode opening was closed with help of a septum and sealed with silicones to prevent gas exchange. Later, the MFC reactors were transferred to a temperature controlled incubator at 30 ± 1 °C; however, this operation temperature was changed during testing the effect of temperature (20 °C, 30 °C and 35 °C). After operating for 2 month (2–3 reloading) with wastewater a piece of (1 cm × 2.8 cm) anode electrode was obtained by cutting anode electrode of all MFC reactor at the same time (end of cycle), and the biomass was extracted. The total extraction of biomass from anode electrode was done in an anaerobic glove box, which was purged continuously with pure nitrogen gas. The anolyte (anodic solution) suspended biomass (planktonic biomass) was also collected and measured by using protein assay to determine microbial growth (planktonic biomass). Then reactors medium was changed to GM media and repeated this process of biomass isolation and estimation in end of operation cycle as shown above.

Analyses

The protein concentrations of bacterial biomass on MFC anode electrode and in anolyte were measured using bicinchoninic acid (BCA) method as previously described [1]. The bacterial biomass obtained from both anode electrode and anolyte (centrifuged) were used for measurement of protein. The extraction of protein from bacterial cell was carried out as described previously [9,10]. During biomass collection from anode electrode, a certain piece of anode (1 cm × 2.8 cm) was cut with help of a sterilized scissors and placed into a conical tube having 15 ml of 0.2 N NaOH. Then this conical tube was kept at 4 °C in refrigerator about 1 h with mixing for every 15 min time interval using a vortex shaker. After that the anode piece was further rinsed with ultrapure water (15 ml) to collect residual biomass on anode electrode. Then this obtained 30 ml of biomass solution at 0.1 N NaOH was treated with three freeze-thaw cycles (frozen at −20 °C and then thawed at 90 °C for 10 min) for extraction protein from bacterial biomass. In the same way, the planktonic biomass obtained by centrifuging (at 4500 rpm for 10 min) anolyte was combined with 1 ml of 0.1 N NaOH, and then prone to the freeze-thaw cycles for extraction of protein from planktonic biomass (anolyte). The SCOD concentration was measured by filtering the samples using syringe filter (Minisart RC 25, Germany, 0.20 μm) and then measured by HUMAS COD-M kit (50–1500 mg/l).

Measurements and calculations

MFC cell voltage was measured and saved for every 5 min using a data acquisition system (National Instruments 9205,