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Comparative Study of Microbial Fuel Cell for Electricity Generation by Enriched Exoelectron Generating Bacteria from Environmental Samples

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ABSTRACT

Microbial Fuel Cells (MFCs) have attracted considerable attention over the last decade as a sustainable technology for electricity generation. Although, the concept of electricity production from bacteria was conceived nearly a century ago, only recently the technology has been sufficiently improved to make it useful as a method for energy generation. Performances of constructed MFCs with different enriched environmental samples were investigated under batch mode of operation after 24 h of incubation using different cathodic electrolytes and electrodes in mediator-less and with methylene blue as mediator. The maximum power density obtained was 48.85 mW m^{-2} with graphite rod and 42.59 mW m^{-2} with copper electrode and current density obtained was 108.57 mA m^{-2} with graphite rod and 88.01 mA m^{-2} with copper electrode all obtained with $KMnO_4$ solution.

Key words: Microbial fuel cells, anaerobic bacteria, exoelectron generating bacteria, catholytes, mediator, power density

INTRODUCTION

An increasing need for new energy sources due to the concerns of the limited availability of fossil fuels has motivated the development of alternate sources of energy. Microorganisms are potential sources for development of such alternative fuels from biomass which are also called as biofuels. Biofuels such as biodiesel (Ibeto *et al*., 2011; Igwenyi *et al*., 2011; Motojesi *et al*., 2011; Alkabbashi *et al*., 2009; Wan Omar *et al*., 2009), bioalcohols (Asad-ur-Rehman *et al*., 2008), biohydrogen (Nazlina *et al*., 2009; Yusoff *et al*., 2009) have been well described in literature.

Apart from biofuels, microorganisms have also been effectively used as a source of electricity by the use of Microbial fuel cell technology (Ho *et al*., 2009; Lim *et al*., 2010). Microbial fuel cells are the devices which convert energy associated with organic substrates into electricity using microorganisms as catalysts. In normal fuel cells, chemical oxidants oxidize the fuel and generated electrons travel across anode to cathode where an electron acceptor gets reduced thereby generating an electric current. Similarly in a microbial fuel cell, the oxidation of the fuel (or substrate) is brought about as a result of microbial metabolism. The primary mechanisms for electrochemically active microorganisms which are the key biocatalysts involved in electricity generation in MFCs, to transfer electrons to the electrodes, metabolic or physiological characteristics and the fundamentals of the anodic or cathodic reactions are well understood (Schroder, 2007).

MFC-microbial communities have been known to degrade a wide range of substrates and pollutants. MFCs have operational and functional advantage over other technologies used for generating current from organic material. Firstly, direct conversion of substrate to electricity enables high conversion efficiency. Secondly, MFCs operate efficiently at ambient and even at low temperature, thus distinguishing them from current bio-energy processes. Thirdly, they don't require gas treatment, as off gasses from MFCs are enriched with CO_2 with no useful energy content. Fourthly, they don't require energy input for aeration and lastly MFC technology has widespread application in locations lacking electrical infrastructure.

Many anaerobic bacteria (DiChristina *et al*., 2002; Lovley, 2006) are known to produce electricity when supplied with an efficient electron acceptor or some mediators which assist microbes for electron transfer. These bacteria are known to be electrochemically active and are usually known as exoelectrogens.

Exoelectrogens are mostly metal reducing anaerobic bacteria which utilize metal ions as substrate for transfer of electrons from their surface to outside the bacterial cell. Many anaerobes can only transfer electrons to soluble compounds such as nitrate or sulphate that can diffuse across the cell membrane and into the cell. Exoelectrogenic bacteria are distinguished from these anaerobes by their ability to directly transport electrons outside of the cell which permits them to function in an MFC.

Microbial Fuel Cells (MFC) are electrochemical devices that convert the chemical energy contained in organic matter into electricity by means of the catalytic (metabolic) activity of living microorganisms (Mathuriya and Sharma, 2008; Bennetto, 1990; Kim *et al*., 2002; Rosenbaum *et al*., 2007). Microorganisms oxidize the substrate and produce electrons and protons in the anode chamber. Electrons, collected on the anode, are transported to cathode by external circuit and protons are transferred through the membrane internally. Thus, potential difference is produced between anode chamber and cathode chamber due to dissimilar liquid solutions. Electrons and protons are consumed in the cathode chamber by reducing oxygen, usually from water. MFCs can be classified into two types. One type generates electricity from the addition of artificial electron shuttles (mediators) to accomplish electron transfer to the electrode. The other type does not require these additions of exogenous chemicals and can be loosely defined as a mediator-less MFC. Mediators can divert electrons from the respiratory chain by entering the outer cell membrane, becoming reduced and then leaving in a reduced state to shuttle the electron to the electrode. Bacteria present in mediator-less MFCs have electrochemically active redox enzymes on their outer membranes that transfer the electrons to external materials and therefore, do not require exogenous chemicals to accomplish electron transfer to the electrode.

In the present study two-compartment MFCs have been constructed with different enriched environmental samples and their efficiency have been analysed for power generation.

MATERIALS AND METHODS

Medium composition and inoculum source: The four collected samples were inoculated in enrichment medium for culturing anaerobic bacteria with composition peptone (15 g L^{-1}), yeast extract (5 g L^{-1}), D-glucose(5.5 g L^{-1}), sodium chloride (2.5 g L^{-1}), cysteine hydrochloride (0.5 g L^{-1}) and agar (0.75 g L^{-1}). Cysteine hydrochloride is a common reducing agent used to depress the redox potential of the medium which is usually required for the growth of anaerobes. Addition of agar increased the viscosity of medium which reduced the diffusion of atmospheric oxygen into medium. The above cited four samples contained two samples each of soil and water. Soil samples were

collected from Mahamaya Ispat industry and Abhishek industry. Both of these are iron-ore industries located in Raipur. These sites were selected because of the probability of presence of iron reducing bacteria or exoelectrogens in these sites. Water samples were collected from two sources, first, from industrial waste water line of Urla industrial region and second from sewage tank of Amleshwar locality, both from Raipur.

Cultivation: One percent of each of the four samples was inoculated in 20 mL enrichment medium described above and incubated at room temperature without agitation under anaerobic conditions. The anaerobic conditions were maintained by sealing the culture tube after flushing with nitrogen gas and applying a layer (approx. 2 cm) of autoclaved oil over the surface of medium. This ensured anaerobic conditions and therefore, allowed the growth of only anaerobes. At the end of 24 h of incubation the enriched cultures were analysed for bioelectricity generation.

Microbial fuel cell construction and operation: Various forms of two cell compartment electrochemical cell may be devised to demonstrate microbial electricity generation and the one described here is one of them. A Microbial Fuel Cell (MFC) constructed in present study is a two-compartment structure divided by a salt bridge. The electrons available through the metabolism of the electron donors by microorganisms are transferred to the anode of the fuel cell and then to the cathode through the circuit, where they reduce the oxidant (Allen and Bennetto, 1993), consuming protons available through the membrane from the anode.

MFCs were constructed using two glass test tubes (35 mL capacity) connected with a salt bridge (5 mm diameter). Salt bridge was constructed using plastic U-tube filled with KNO_3 (saturated) and agar (20 g L^{-1}). Three different electrodes made up of graphite rod (14.828 sq. cm), carbon paper (34.2 sq. cm) and copper plate (15.68 sq. cm) were used. Copper wires of resistance 3.4×10^{-3} Ω m^{-1} were used for connecting circuits. The electrodes were soaked in phosphate buffer (50 mM) before placing in MFC. Different cathodic electrolytes were used to compare the performance of MFC. The cathodic electrolytes used were tap water, NaCl (10 g L^{-1}) with tap water, NaCl (10 g L^{-1}) with aerated tap water and $KMnO_4$ (0.2 g L^{-1}) with tap water. The enriched cultures were directly taken as anode. Methylene blue (0.1 mM) was used as mediator to study the performance of MFC. Potential (V volt) and current (I amp) were measured using a digital multimeter (Mastech, M-830BZ).

Calculation: Current density of MFC was calculated using i (mA m^{-2}) I/A where I is the current measured (mA) and A is the geometric surface area of anode (m^2). The power density of the MFC was calculated using formula: P (mW m^{-2}) = IV where I is the current density and V is the voltage measured (mV).

RESULTS AND DISCUSSION

Enrichment of anaerobic exoelectron generating bacteria has been reported by Bond and Lovley (2003), Kim *et al.* (2002). In the present study glucose was used in the medium as carbon source in construction of fuel cell. Natural micro-flora of Palm Oil Mill Effluent (POME) sludge was also grown in dual-chamber Microbial Fuel Cells (MFC) to produce electricity by providing glucose at different concentration (Lim *et al.*, 2010). Scott and Murano (2007) have also constructed the carbohydrate utilizing MFC. The comparative production of electricity from all four different samples with all variables has been shown in Table 1 and 2. Similar electricity generation from

Table 1: Measure of voltage, current, current density and power density in all samples with graphite as electrode

Sample	Cathodic electrolyte	Voltage (mV)	Current (mA)	Current density (mA m^{-2})	Power density (mW m^{-2})
Soil sample from	Tap water	418.0	0.132	89.02	37.21
Mahamaya Ispat	Tap water with NaCl	116.0	0.057	38.44	4.46
industry	Aerated tap water with NaCl	134.0	0.041	27.65	3.71
	Tap water with $KMnO_4$	440.0	0.148	99.81	43.92
	Tap water with $KMnO_4$ and methylene blue	305.0	0.141	95.09	29.00
Soil sample from	Tap water	376.0	0.120	80.92	30.43
Abhishek industry	Tap water with NaCl	22.0	0.009	6.06	0.133
	Aerated tap water with NaCl	86.4	0.040	26.97	2.33
	Tap water with $KMnO_4$	432.0	0.143	96.44	41.66
	Tap water with $KMnO_4$ and methylene blue	265.0	0.121	81.60	21.62
Water sample from	Tap water	248.0	0.096	64.74	16.05
industrial waste	Tap water with NaCl	88.5	0.038	25.63	2.27
Water line of Urla	Aerated tap water with NaCl	74.6	0.035	23.60	1.76
	Tap water with $KMnO_4$	392.0	0.146	98.46	38.59
	Tap water with $KMnO_4$ and methylene blue	234.0	0.091	61.37	14.36
Water sample from	Tap water	172.4	0.075	50.57	8.72
Amleshwar locality	Tap water with NaCl	112.4	0.055	37.09	4.17
	Aerated tap water with NaCl	19.3	0.010	67.43	1.30
	Tap water with $KMnO_4$	450.0	0.161	108.57	48.85
	Tap water with $KMnO_4$ and methylene blue	167.0	0.064	43.16	7.21

Table 2: Measure of voltage, current, current density and power density in all four samples with copper as electrode

Sample	Cathodic electrolyte	Voltage (mV)	Current (mA)	Current density (mA m^{-2})	Power density (mW m^{-2})
Soil sample from Mahamaya	Tap water	284	0.048	30.61	8.69
Ispat industry	Tap water with NaCl	312	0.090	57.39	17.91
	Aerated tap water with NaCl	67	0.029	18.49	1.24
	Tap water with $KMnO_4$	474	0.123	78.44	37.18
	Tap water with $KMnO_4$ and methylene blue	471	0.138	88.01	41.45
Soil sample from Abhishek	Tap water	215	0.056	35.71	3.68
industry	Tap water with NaCl	53	0.018	11.47	0.61
	Aerated tap water with NaCl	110	0.046	29.33	3.23
	Tap water with $KMnO_4$	431	0.125	79.71	34.35
	Tap water with $KMnO_4$ and methylene blue	504	0.130	82.91	41.78
Water sample from industrial	Tap water	224	0.048	30.61	6.86
waste water line of Urla	Tap water with NaCl	51	0.015	9.56	0.49
	Aerated tap water with NaCl	38	0.017	10.84	0.41
	Tap water with $KMnO_4$	506	0.132	84.18	42.59
	Tap water with $KMnO_4$ and methylene blue	396	0.133	84.82	33.58
Water sample from Amleshwar	Tap water	120	0.027	17.21	2.06
locality	Tap water with NaCl	233	0.100	63.77	14.86
	Aerated tap water with NaCl	102	0.041	26.15	2.67
	Tap water with $KMnO_4$	399	0.113	72.06	28.75
	Tap water with $KMnO_4$ and methylene blue	358	0.121	77.16	27.62

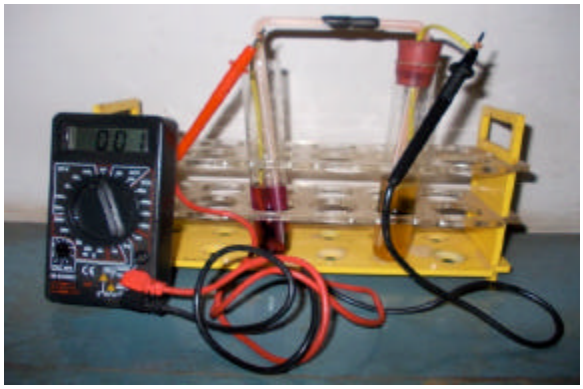

Fig. 1: Microbial fuel cell

waste water sample have been done by Patil *et al.* (2011). Presence of mediator (methylene blue) does not execute the enhanced current production. Table 1 and 2 show that cathodic electrolyte has significant effects on power density. In all the samples tap water with $KMnO_4$ resulted in maximum current density (Table 1, 2). Jadhav and Ghangrekar (2008) found the effect of catholyte for improving the performance in fuel cell.

The maximum current density was found with graphite electrode water sample from Amleshwar locality (108.57 mA m^{-2}) and with carbon electrode from water sample from industrial waste water line of urla (84.82 mA m^{-2}). The enriched exoelectron generating bacteria demonstrates the electricity generation which can be further improved and may have other application too as reported by Kim *et al.* (2008).

Figure 1 shows the construction of microbial fuel cell using the enriched anaerobic bacteria from soil and water samples for exoelectron generation and electricity production.

CONCLUSION

The maximum power density obtained was 48.85 mW m^{-2} with graphite rod and 42.59 mW m^{-2} with copper electrode and current density obtained was 108.57 mA m^{-2} with graphite rod and 88.01 mA m^{-2} with copper electrode all obtained with $KMnO_4$ solution.

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