Dual Chambered Microbial Fuel Cell For Bioelectricity Generation From Environmental Samples

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DOI: 10.29322/IJSRP.9.05.2019.p8996
http://dx.doi.org/10.29322/IJSRP.9.05.2019.p8996

Abstract-. Microbial Fuel Cell(MFC) is an emerging technology which under anaerobic conditions, converts chemical energy intheorganicsubstratesto electrical energy through catalytic reactions of microorganisms. This study is aimed to generate bioelectricity using a fabricated MFC with low cost anode and cathode materials without any toxic mediators. We devised MFC using Plain Graphite plates as electrodes. The cathodic electrolytes used were distilled water with NaCl whereas anodic compartments were inoculated with enriched bacterial culture. Anodic and Cathodic chambers were separated by a Proton Exchange Membrane. Current and Voltage measurements were recorded using a digital Multimeter. The performance of MFC was analysed using Polarization Curves. Maximum Power and Current densities obtained were 91,509.7mW.cm^{-2} and 46.88mA.cm^{-2} respectively. This experimental research was targeted to understand the capability of mediator-less dual chambered microbial fuel cell as electricity production unit.

Index Terms- Anaerobic conditions, Mediator-less MFC, Graphite plates, Electricity

I. Introduction

The International Energy Agency (IEA) in 2013 estimated World energy consumption as 1.575×10^{17} kWh per year. Fossil fuels have supported the industrialization and economic growth of countries during the past century, but they cannot indefinitely sustain a global economy. The use of Fossil fuels leads to negative environmental impacts. Therefore, newer approaches like using fuel cells have emerged as a renewable energy sources that produces sufficient energy while at the same time reduces environmental damage. Among different kinds of fuel cells, Microbial Fuel Cells defined as devices which directly converts microbial metabolism into electricity have attracted researcher's attention (Allen, R.M. et al.,1993). The working principle of MFCs is based on the tenets of microbial physiology coupled with electrochemistry. MFCs are the major type of bio electrochemical systems which convert biomass spontaneously into electricity through the metabolic activity of the microorganisms. MFC technology has been found as a potential technology for electricity generation.

While the first observation of electrical current generated by bacteria is generally credited to Potter in 1911, after William Grove who built the first fuel cell in 1839, very few practical advances were achieved in this field even 55 years later (Lewis. K, 1966). The breakthrough in MFCs occurred in 1999 when it was recognized that mediators need not to be added. The essential physical components of the MFC are the anode, cathode and electrolyte. MFCs are typically designed as a two-chamber system (shown in Fig 1) with the bacteria in the anode chamber separated from cathode chamber by a polymeric Proton Exchange Membrane, which often separates the liquid in the two chambers while allowing protons to pass between the chambers. Microorganisms catalyze the oxidation of substrates releasing electrons and protons in the anode chamber. Electrons, collected on the anode, are transported to cathode by external circuit to create current and protons are transferred through the membrane internally, to sustain the current. Thus, due to dissimilar liquid solutions, a potential difference is produced between anode chamber and cathode chamber. Typically, the electrons and protons react with oxygen at the cathode, to form water. The defining characteristics of an MFC includes microbially catalyzed electron liberation at the anode and subsequent electron consumption at the cathode are sustainable.Principally, the output power depends on the rate of substrate degradation, the rate of electron transfer from the bacteria to the electrode, the circuit resistance and the proton mass transfer in the liquid. Since most microbial cells are electrochemically inactive, rapid electron transfer from a microorganism to an electrode needs an electron mediators like thionine, humic acid and many more. Thus, depending on the presence or absence of mediators, 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 thus can be defined as mediator-less MFCs. Some anaerobic bacteria are even able to transfer electrons extracellularly to insoluble, solid-state electron acceptors such as electrodes (DiChristina et al., 2002).

Applications of MFCs:

The electricity produced by MFCs can be used for powering other technologies, such as biologically inspired robots, some remote devices. In addition, the voltage generated by MFCs can be used on microbial electrolysis cells (MECs). MFCs are especially suitable for powering small telemetry systems and wireless sensors that have only low power requirements to transmit signals such as temperature to receivers in remote locations (Ieropoulos et al., 2005). MFCs provide a renewable hydrogen source that can contribute to the overall hydrogen demand in a hydrogen economy.MFCs can potentially produce about 8–9 mol H_2/mol glucose compared to the typical 4 mol H_2/mol glucose achieved in conventional fermentation (Liu, H., et al., 2005). MFCs were considered to be used in treating sanitary waste, food processing wastewater, swine wastewater(Zhuwei Du, Haoran Li, 2007). MFC type of BOD sensor is used as a sensor for pollutant analysis and in situ process monitoring and control (Picioreanu. C, et al., 2007). Microbial communities in MFCs can function as tool for bioremediation (Jeffrey M. Morris and Song Jin., 2007). The main objective of the present study was to design an MFC which is capable of generating power simultaneously while employing low-cost materials without using toxic mediators. The fabricated MFC system (graphite electrode without any coating) was evaluated at ambient conditions using anaerobic mixed consortia and glucose as the sole source of electron. The current produced by an MFC is typically calculated in the laboratory by monitoring the voltage drop across the resistor using a multimeter.

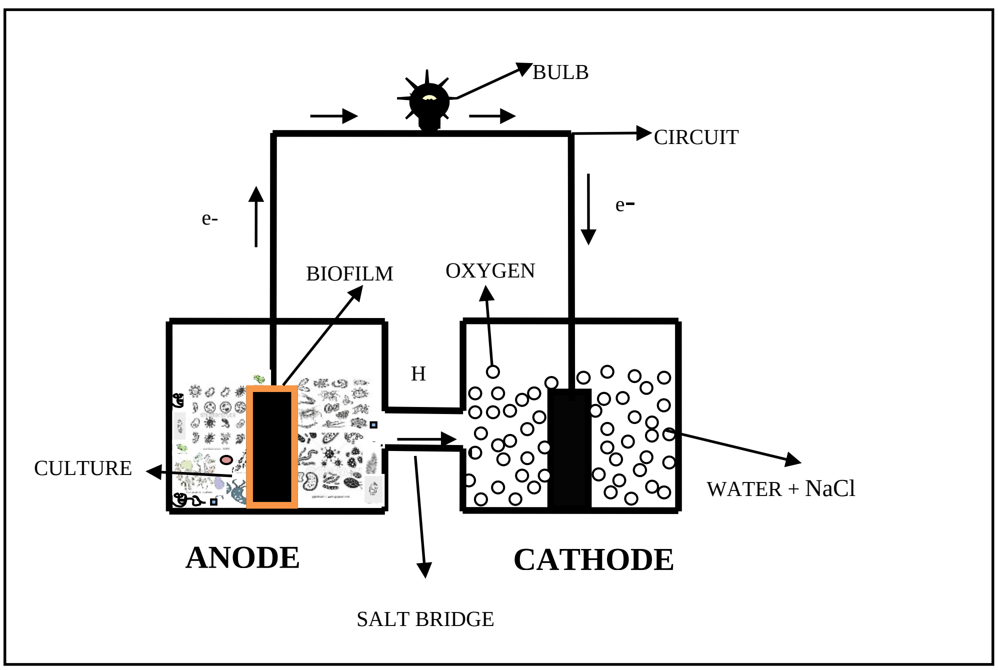


Fig 1: Schematic diagram of a typical two-chamber MFC

MATERIALS AND METHODS

Medium Composition and inoculum source-

Samples of coal were collected to isolate electrochemically active bacteria. The collected samples were inoculated in enrichment medium (Anaerobic media- Tryptone – 20gm/l, Dextrose – 10gm/l, Sodium Chloride – 5gm/l, Sodium thioglycolate – 2gm/l, Sodium formaldehyde sulfoxylate – 1gm/l, Methylene blue – 0.002gm/l, Final pH at 25°c – 7.2 ±2). This medium contains Sodium thioglycolate and Sodium formaldehyde sulfoxylate that provide adequate anaerobiosis which is indicated by methylene blue present in the medium, which gives blue colour to medium in the presence of oxygen. Dextrose provide essential nutrients while Sodium Chloride maintains Osmotic equilibrium. For biochemical characteristic analysis of isolated microorganisms, different media and reagents were used (Paul C. Schreckenberger, et al, 1974).

Cultivation-

One percent of the sample (which was diluted serially) was inoculated into autoclaved enrichment medium and incubated at 37°C without agitation under anaerobic conditions. The anaerobic conditions were maintained by applying a layer (approx. 2cm) of autoclaved oil over the surface of medium and sealed with parafilm wax. This ensures anaerobic conditions and therefore, allows the growth of only anaerobes. At the end of 72hr of incubation, the enriched cultures were analysed for bioelectricity generation.

MFC Fabrication and Operation:

The performance of MFC largely depends on the reactor design and electrode materials. 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. The design criteria of dual chambered fuel cell are shown in Table 1. 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, consuming protons available through the membrane from the anode (Allen, R.M., et al., 1993). Two chambers were selected in order to avoid mixing solutions of electron acceptors and electron donors and thus loss of electrons.

H – type two-chamber MFCs was constructed as described by Oh et al. Two polycarbonate bottles (1000ml) were linked with PVC pipe of 5cm length, which serves as salt bridge. The salt bridge was filled with boiled and cooled Sodium Chloride (15%) solution containing 5% agar. The salt bridge was fixed to the bottles with the aid of epoxy adhesive. The culture inoculum of bacterial strains was used as anolyte without any pre-treatment and distilled water with added NaCl was used as a catholyte in MFC setup. Plain graphite plates (5 – 4cm; 0.3mm thick; 45.4cm^2surface area) were used as electrodes. The electrodes were positioned at a distance of 2cm on either side of the PEM. Circuit connections were set with the copper wires fixed into the drilled holes of the electrodes and sealed with epoxy resin to avoid corrosion of copper wires. The MFC was sterilized by thorough rinsing with Ethanol (70% v/v) and UV irradiated for 30min. The sterilized MFC chambers were filled with respective electrolytes and kept at room temperature for one day. Electricity generation was recorded using a multidigital meter for 21days.

Table 1: Design criteria of Dual chambered mediator-less Microbial Fuel Cell

REACTOR CONFIGURATION	DUAL CHAMBER
Anode chamber	suspended growth
Anode inoculum	Mixed anaerobic Consortia
Mediator-anode	Mediator less
Working volume of anode and cathode	500ml
Electrodes	Graphite plates
Surface area of electrodes	45.4cm²
Membrane	PEM (salt bridge)
Operating temperature	Room temperature

Calculations:

Current density of MFC was calculated using formula:
$i\ (mA.cm^{-2}) = I/A$
where, ‘I’ is the current measured (mA) and ‘A’ is the geometric surface area of anode (cm^2).
The Power density of the MFC was calculated using formula:
$P\ (mW.cm^{-2}) = iV$
where, ‘i’ is the current density and ‘V’ is the Voltage measured (mV).

RESULTS

Enrichment of anaerobic exoelectron generating bacteria has been reported earlier.A layer of biofilm is formed on the surface of the anodic graphite plate. This biofilm formation helps in transfer of exoelectrons to the graphite plate. The present study shows that the microbial inhabitant of environmental sample has electrogenic potential and they can manifest this potential if they are properly re-inoculated into their habitat. This leads to the production of bioelectricity in the fabricated MFC.

A total of seven distinct bacterial isolates were isolated from chosen environmental samples (coal) and their biochemical characteristic analysis (using tests like IMVIC, gelatine, nitrate reduction test, urease, catalase, oxidase, Esculin hydrolysis and Sugar fermentation) were carried out and the results are shown in Table 2. MFC constructed by using 1000ml PVC bottles and connected with 5cm long and 5cm diameter salt bridge was used to generate electricity by microbial cell suspension cultures. Power generation in mediator-less microbial fuel cell was investigated using a digital multimeter. The increase in production of electricity from MFC is shown in Table 3.

ISSN 2250-3153

Table 2: Results of Biochemical characteristic analysis of isolated bacteria

Organism expected to be / Tests For identification		*Bacillus* sps.	*Acidothiobacillus* sps.	*Pseudomonas* sps.	*E. coli*	*Staphylococcus* sps.	*Clostridium* sps.	*Propionibacterium* sps.
Gram's staining		Gram +ve	Gram -ve	Gram -ve	Gram -ve	Gram +ve	Gram +ve	Gram +ve
Shape		Rods	Rods	Rods	Rods	Cocci	Rods	Rods
Motility		Motile	Motile	Motile	Motile	Non-motile	Motile	Non-motile
Capsulated		-	-	+	+	+	-	-
Sporulation		+	-	-	-	-	+	-
Catalase		+	-	+	+	+	-	+
Oxidase		-	+	+	-	-	-	-
Urease		+	-	-	-	+	-	-
Indole		+	-	-	+	-	-	+
MR		+	-	-	+	+	+	-
VP		+	+	-	-	+	-	-
Citrate utilization		+	+	+	-	+	+	-
Nitrate reduction		-	+	-	+	+	-	-
Gelatin hydrolysis		+	-	+	-	+	+	-
Esculin hydrolysis		+	+	-	+	-	+	+
Starch hydrolysis		+	-	-	-	-	-	-
Sugar fermentation tests	**G**	+	+	-	+	+	+	+
	L	-	+	-	+	+	+	+
	M	-	+	-	-	+	-	-
	S	+	+	-	+	+	+	-

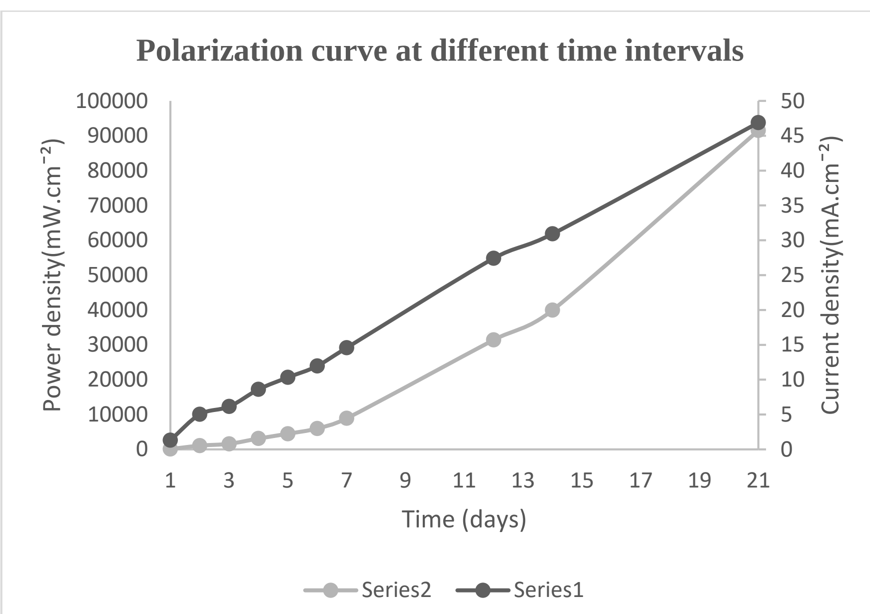
Where, G = Glucose, L = Lactose, M = Mannose, S = Sucrose.
'+' indicates – the reaction is positive for the test and '- 'indicates – the reaction is negative for the test.

Table 3: Measure of Voltage, Current, Current density and Power density for a period of 21days with graphite as electrode

S.No.	Day	Current measured in milliamperes(mA)	Voltage measured in millivolts(mV)	Current density(i) in $mA.cm^{-2}$	Power density(P) in $mW.cm^{-2}$
1	Day 1	59.7	55	1.31	72.05
2	Day 2	228.2	210	5.02	1054.2
3	Day 3	279.2	257	6.14	1577.9
4	Day 4	391.2	360	8.61	3101.7
5	Day 5	469	432	10.33	4462.5
6	Day 6	543	500	11.96	5980
7	Day 7	662	610	14.58	8893.8
8	Day 12	1245.3	1146	27.42	31423.3
9	Day 14	1403.9	1292	30.92	39948.6
10	Day 21	2128.7	1952	46.88	91509.7

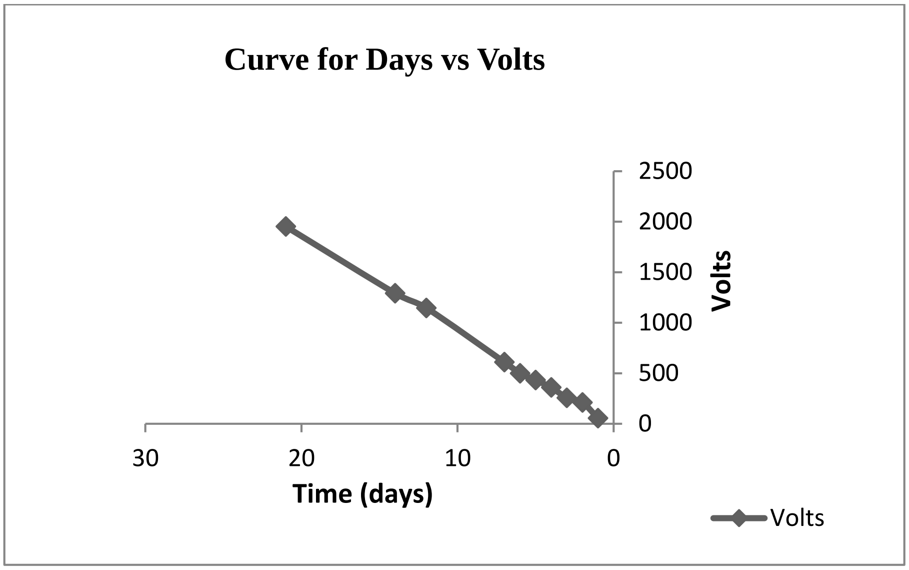
As shown in Fig.2 and Fig.3, there is an increase in Power production with increasing time period. This is due to the formation of bacterial biofilm on the graphite surface in anode chamber. About two days after the beginning of the MFC operation, the current and power densities increased. Probably, this period was required by the microorganisms to adapt to the culture medium, to begin to adhere to the graphite plate, and to produce the biofilm. The Power density was directly dependent on the biofilm development. The spreading property of biofilm is especially useful for MFC efficiency because it means that the number of electrons transferred to the anode is directly proportional to the anode's surface area given minimal loss of electrons due to close proximity between the biofilm and the anode (Kim BH, et al., 2003). The maximum Current density and Power density was $46.88 mA.cm^{-2}$ and $91,509.7 mW.cm^{-2}$.

Fig. 2: Polarization curve to show Power generation in MFC



Where, series 1 represents Current density and series 2 represents Power density

Fig.3: A curve showing an increase in volts in MFC



CONCLUSION

Microscopic, Morphological and Biochemical characteristic analysis revealed the presence of seven different microorganisms which include aerobes, anaerobes and facultative anaerobes. The results revealed the feasibility of bioelectricity generation using a fabricated MFC with low cost anode materials (non-coated plain graphite electrodes), without any toxic mediators. It can hence be stated that the potential electrogenic bacteria can be easily procured from environment and deployed in MFCs for an eco-friendly and economically viable method of electricity generation. Maximum Current and Power densities obtained using anaerobic consortia in this experiment were 46.88mA.cm^{-2} and 91,509.7mW.cm^{-2}. Although MFCs are a promising technology for renewable energy production, they face several challenges, as well.

ACKNOWLEDGMENTS

The authors gratefully acknowledge the Department of Microbiology, Osmania University for providing the necessary facilities to carry out the present study.

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