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Bioelectricity Production in a Double Dual Chambered Microbial Fuel Cell through Anaerobic Treatment of Distillery Wastewater

Shambhavi R S[1], Udayashankara T H[2], Pallavi C K[3]
[1](Assistant Professor, Department of Civil Engineering,Jyothy Institute of Technology, Bengaluru-560062, Karnataka, India)
[2](Professor, Department of Environmental Engineering, Sri Jayachamarajendra College of Engineering, Mysuru-570006, Karnataka, India)
[3](M. Tech Scholar, Department of Environmental Engineering, Sri Jayachamarajendra College of Engineering, Mysuru-570006, Karnataka, India)

Abstract: Microbial Fuel Cells (MFC) are bio-ethanol devices that harness the natural metabolisms of microbes to produce electrical power. Within the MFC, microbes munch up the sugars and othernutrients in theirsurroundingenvironment and release a portion of the energycontainedwithin the food in the form of electricity. MFCs are attractive for wastewatertreatment, becausetheycouldallow for harvestingenergyfromwastewater for producingelectricity. The goal of thisprojectwas to build Double Dual chamberedMFC(DDMFC) thatharvestelectricity and producereclaimed water fromwastewater (distillery) i.e., treatment of distillerywastewater. MFCs were constructed from cheap alternatives such as agar salt bridge to traditionally used expensive Nafion membranes and also without using costly mediators. Optimisation of DDMFC wascarried out by varying the volume of the wastewatersample. In thisstudy DDMFC and Double chamberedMFC (DMFC) werecompared for the generation of electricity. DDMFC was efficient and maximum voltage of 2.010V wasproduced and COD removalwas 93%. MFCprovide a method of addingwastewater to the list of renewableenergy sources.

Keywords: Microbial fuel cell, distillery wastewater, voltage, COD

I. INTRODUCTION

The global energy demand increases the difficulty in sustained supply and the associated problems of pollution and global warming are acting as a major impetus for research into alternative renewable energy technologies. More recently, it has been reported that microorganisms can also convert organic matter into electricity using MFC. The MFC is a new technology in which microorganisms produce electricity directly from renewable biodegradable material. MFCs provide a method of adding wastewater to the list of renewable energy source. To apply the MFC in wastewater treatment system, several advantages have been stated including 1) production of a useful product in the form of electricity; 2) lack of need for aeration; 3) reduced solids production and 4) potential for odor control [1]. Apart from high organic content, distillery wastewater also contains nutrients in the form of nitrogen, phosphorous and potassium that can lead to eutrophication. The organic load of distillery spillages is characterized by a high content of glycerol and organic acids (lactic, tartaric). Various anaerobic bacteria ferment these compounds and generate products such as volatile fatty acids[2]. The main objective of the present work is bioelectricity production and simultaneously treating the distillery wastewater using DDMFC. In this study efficiency of DDMFC was evaluated by employing low cost materials. A comparative study was also done between DMFC and DDMFC. Optimization of MFC was carried out by identifying rate limiting factors

Table 1: Power generation rates reported in the literature

Sl. No.	Wastewater used	Inoculum	MFC Configuration	Current, mA	Power density,	References
1.	Brewery	Glucose	Two chamber	10.89	NA	[3]
2.	Paneer whey	*Klebsiellapneumonae*	Two chamber	0.41	NA	[4]
3.	Domestic	NA	Flat plate Type MFC	NA	72 mW/m^2	[5]
4.	Synthetic	Septic tank sludge	Membraneless MFC	0.175	6.73 mW/m^2	[6]
5.	Distillery	Sewage	Two chamber	0.34	17.76 mW/m^2	[7]
6.	Domestic	Bacterial culture	Tubular Type MFC	NA	48 W/m^3	[8]
7.	Rice mill	Anaerobic sludge	Two chamber	1.07	15.57 mW/m^2	[9]
8.	Abattoir	NA	Stacked double two chamber	NA	20.17 mW/m^2	[10]

9.	Domestic	NA	Salt bridge immersed air cathode MFC	0.256	NA	[11]
10.	Synthetic	Anaerobic sludge	Up flow type NFC	NA	315 W/m^3	[12]

II. MATERIALS AND METHODOLOGY

An MFC requires much the same components as any other fuel cell however there are some drastic differences. Most obvious is the requirement for bacteria which is not an addition to any other type of fuel cell. Where most other fuel cells incorporate chemicals to achieve their electricity producing reactions, a MFC requires a form of organic matter (substrate) to maintain the bacteria also providing the means of generation. Low-cost substitutes for each MFC component were identified as follows

2.1 MFC reactors: The DDMFC for use in laboratory scale experiments was set-up using cylindrical type plastic containers of size (15 cm x 15 cm, volume=2.5 L). Four such containers were used to form the DDMFC and were connected in series. Easily available and inexpensive plastic containers were used instead of borosil bottles which are expensive.

2.2 Electrode materials: Both anode and cathode electrodes were made of graphite rods. The length and diameter of graphite rods were 15 cm and 1.5 cm respectively. Both the electrodes were positioned at a distance of 6 cm from each other because maximum electricity is generated when electrodes are placed close to each other. Pretreatment was not provided for the electrode materials.

2.3 Membrane: Membranes used are generally veryexpensive, have high minimum orders or incur large freight charges as they are onlymanufactured overseas. So agar-salt bridge was used in this study. A water solution containing concentrations of 1 M KCland 2-3% agar was allowed to boil for nearly 3 minutes. The hot solution was poured into PVC pipe of 30 cm length and 0.6 cm internal diameter. The setup was thereafter allowed to cool for nearly 2 hours. The salt bridges were thus ready for use.

2.4 Catalyst/catholyte: The cathode chamber is where protons and electrons recombine and reduce with an electron acceptor. A common electrode acceptor is oxygen due to its abundance in air. When oxygen is used however the reaction is very slow and leads to a high over potential. To overcome this electrode over potential, laboratory based MFC systems commonly use potassium permanganate as an electron acceptor. The use of potassium permanganate leads to a low over potential of the graphite cathode, and allows the MFC to work close to open circuit potential. Most MFC's use platinum as the catalyst however this is extremely expensive. So potassium permanganate was used successfully in this study with results comparable to those achieved with platinum. To increase the DO content, aerators were used for continuous aeration of cathode chamber.

2.5 Microorganism: In the present study, *Saccharomyces Cereviceae* was used as microorganism (biocatalyst) which was already present in wastewater without using mediators since yeast is an axenic microorganism requires mediators to transport electrons to the electrode. The yeast will convert sugar components in the wastewater into CO_2, wherein the intermediate process will be release electron generating electricity in MFC system. Yeasts have recently been used to generate electricity in microbial fuel cells, and to produce ethanol for the biofuel industry. *SaccharomycesCereviceae* yeasts have been genetically engineered to ferment xylose, one of the major fermentable sugars present in cellulosic biomasses, such as agriculture residues, paper wastes, and wood chips and also in distillery wastewater.

2.6 Substrate: The distillery and sugar wastewaters were used as substrate. Distillery wastewater was considered as the substrate because of its high organic content and sugar wastewater was used as another substrate because it contains simple compounds which can be easily digested by the microorganisms. No any additional nutrients were given for microorganisms except the nutrients present in the distillery and sugar wastewaters.

The distillery wastewater was collected from two different distillery industries.

1. **NSL (Nuziveedu Seeds Limited) Sugars** located at Koppa Village, Maddurtaluk of Mandya District. The sample was collected from the collection tank.

2. **M/S. Coromandel Sugars Ltd** located at S.F.Nos.151, Makavalli Village, K. R. Pet Taluk, Mandya District, Karnataka, which islocated 75 km away from Mandya. Distillery wastewater sample was collected from the collection tank and the sugar wastewater sample was collected from the spray pond.

Table 2: Characteristics of distillery and sugar wastewater

Sl. No.	Parameters	Concentration		
		ICL		NSL
		Sugar	Distillery	
1.	pH	5.7	4.3	3.9
2.	Color	Pale Yellow	Dark Brown	Dark Brown
3.	BOD (mg/L)	1058	39000	66000
4.	COD (mg/L)	1228	44800	118400
5.	Volatile Solids (mg/L)	750	17415	106680
6.	Total Solids (mg/L)	1200	21360	159070
7.	Phosphates (mg/L)	7.5	800	2761
8.	Sulphates (mg/L)	27.3	1925	3375
9.	Nitrates (mg/L)	26	1120	3000
10.	Chlorides (mg/L)	1.99	950	1045
11.	BOD/COD ratio	0.86	0.87	0.55
12.	TS/VS ratio	1.6	1.22	1.49

2.7 Construction:

The DDMFC used for the experiment is as shown in figure. Four plastic containers were used to form the DDMFC connected in series. Agar salt-bridge was used as proton exchange membrane. The electrodes were connected using copper wires and the circuit was made complete. The copper conducts the electron flow generated from the electrode surface across external electrical circuit. In the second chamber of the MFC, there was another solution and another electrode. This electrode, called the cathode would be positively charged and would be the equivalent of the oxygen sink at the end of the electron transport chain, only now it would be external to the biological cell. The solution would be an oxidizing agent that would pick up the electrons at the cathode. Both the cathode chambers were filled with distilled water and 0.2 g of potassium permanganate was added and it was continuously aerated. The anode chamber was filled with distillery wastewater of 1 L volume. The microorganisms and the organic matter present in the wastewater were sufficient for the voltage generation. Performance of DDMFC was evaluated at four different experiments containing 2 cycles per experiment. Each experiment was carried out for 7 days or until there is stabilization. DDMFC was operated in batch mode at ambient temperature (28 ± 2°C) and a pH of 6.5 and 7 was maintained by adding NaOH solution.

2.8 Analyses

Voltage and Current measurement was recorded for every 1 hr using multimeter by connecting 10 Ω external resistance. The influent and effluent COD concentrations, BOD concentrations, pH contents were monitored according to standard methods.

Figure 1: Experimental setup of DDMFC and DMFC

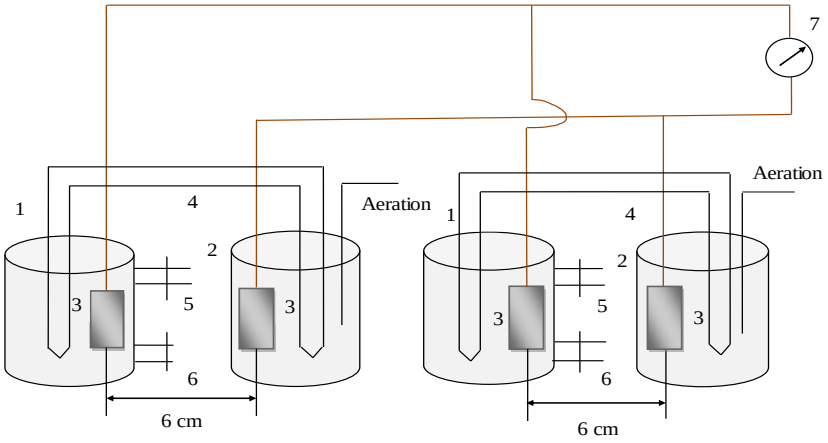

Figure 2: Experimental setup of DDMFC and DMFC

1. Anode chamber
2. Cathode chamber
3. Graphite plate
4. Agar-salt bridge
5. Inlet port
6. Drain port
7. Multimeter

III RESULTS

The fuel cells were operated with distillery wastewater and sugar wastewater. Constant substrate (COD) removal efficiency and voltage output were considered as indicators to assess the stable performance of the MFC. Experimental data showed the feasibility of electricity generation from wastewater treatment. The results obtained in this study are found to be very promising for agar-salt bridge. DO content in the distillery wastewater is nil and it is a favorable condition that is required in the anode chamber of DDMFC. However, the performance and stabilization tendency with respect to power generation was found to be dependent on the substrate load (volume) and redox conditions. In a stack it is the number of cells that contributes to the final voltage, while the surface area of each cell determines the final current. Connecting several fuel cells in series adds the voltages, while one common current flows through all fuel cells. In case several power sources are connected in parallel, the voltage averages and the currents are added. Since a single MFC generate about 0.5V, several single cells should be connected in series to achieve higher voltage.During continuous stack operation voltage reversal will not occur due to high substrate concentrations in both cells and short hydraulic retention times (HRTs). Voltage reversal is produced when voltage in the cells is not matched, which can occur as the result of substrate starvation. Fuel starvation, i.e., an inadequate supply of fuel, is a major cause of cell reversal and can occur during a sudden change of fuel demand such as during start-up or a change of the load.

3.1 Optimization of DDMFC

The DDMFC was more effective in generating voltage than DMFC because when two MFCs are connected in series the voltage will add up and hence the results obtained in this study is in par with the literature. Substrate load had a marked influence on electricity generation. The effects of operational conditions (MFC design, effective assembly of membrane electrode for reducing the internal resistance, providing adequate surface area for the bacterial growth, improving the cathode reaction, selection of bacterial consortium, pH, DO

concentration, temperature, type of substrate, substrate conversion rate, electrode spacing and electrolyte strength) were investigated and optimized for the best performance of a DDMFC.

The only condition to be optimized was the volume of wastewater to be taken for maximum electricity generation. Cycle 6: ICL sugar (distillery wastewater 125 ml + 100 ml sugar wastewater) was found to be optimum because increase in volume of sample decreased the voltage production.To check the performance of MFC, experiment was carried out with distillery wastewater only. Cycle 8 (125 ml distillery wastewater)was found to be optimumin generating maximum voltage but with a longer reaction time. Stabilization is an important part of MFC. The system will be stable only when there is sufficient amount of organic matter in the wastewater. As reaction time increases, the system will be destabilized because decrease in the organic matter present in the wastewater and increase in the microbial concentration and also due to the production of recalcitrant. For re-stabilization of the system, glucose and sugar wastewater were added.

For cycle 5 and 6, glucose was added when there was decrease in the voltage (at 96^{th} hour) but stabilization was not achieved. So, for cycle 8 sugar wastewater was added to check the performance of MFC. After adding sugar wastewater (at 312^{th} hour) the stabilization was achieved after 24 hours and there was increase in voltage generation (1.782 V). The stabilization was achieved with sugar wastewater than with glucose because sugar wastewater not only contains glucose but also sucrose, fructose as sugars through which microorganisms gets more food and thus microorganisms can easily degrade the organic content present in the distillery wastewater.A maximum voltage of 2.012 V for DDMFC and 1.007 V for DMFC; maximum current of 1.873 mA for DDMFC and 0.937 mA for DMFC was generated after 648^{th} hour (figures 4.13 and 4.14). The voltage generation was more after the addition of sugar wastewater because microbial concentration is more in sugar wastewater.

3.2 Effect of Substrate Load on Voltage Generation

The DDMFC was operated with two different distillery industrial wastewaters. Four experiments were carried out, two cycles per experiment. The volume and the combination of the wastewater required in the DDMFC operation are given in the table. The wastewater was diluted four times using distilled water in the cycles 3-8.

Table 3: Volume and combination of the wastewater

Cycle number	Distillery wastewater used	Volume of sample (ml)
1	Chamundi distilleries (distillery wastewater)	1000
2	NSL sugars (distillery wastewater)	750
3	NSL sugars (distillery wastewater)	500
4	NSL sugars (distillery wastewater)	200
5	ICL sugars (distillery wastewater + sugar wastewater)	100 + 100
6	**ICL sugars (distillery wastewater + sugar wastewater)**	**125 + 100**
7	ICL sugars (distillery wastewater + sugar wastewater)	150 + 100
8	**ICL sugars (distillery wastewater)**	**125**

After running the DDMFC (cycle 1), a maximum voltage of 423 mV was achieved with Chamundi Distilleries wastewater. But stabilization was not achieved with this wastewater because high load was applied on the microorganisms (1000 ml). So experiment was carried out in DDMFC with NSL and ICL industrial wastewaters and the volume of the sample was also reduced. Collection and characterization of sample was carried out. 750 ml (cycle 2) of NSL industrial sample was taken and the experiment carried out. About 12.3 mV, 30.32 mV of voltage was generated after 48 and 72 hrs respectively. Stabilization was lost after 96 hrs. This may be due to more loads on the microorganism and the voltage generated was too less for a stacked MFC.

Therefore the volume of sample was further reduced to 500 ml (cycle 3) and 200 ml (cycle 4). 250 ml of sample volume was reduced from cycle 1-3 per cycle, since the voltage generated was not in par with the literature, the reduction was 300 ml from cycle 3 to 4. After loading with the sample, 24 hrs lag phase was given for the microorganisms to get acclimatized to the new environment. And after 24 hrs, for every 1 hour the current and voltage reading was taken because for 30 min interval there was no much increase in voltage generation. The current and voltage readings were noted simultaneously from both DMFC and DDMFC. The same procedure was followed for all the 8 cycles.

The experiment was carried out for 7 days (cycle 2-4, 7) and 6 days (cycle 5, 6) and readings were taken up to 6^{th} day (cycle 2-4, 7) and 5^{th} day (cycle 5, 6).Cycle 8 was operated for 30 days and readings were taken up to 29^{th} day.Since distillery wastewater contains more of polysaccharides, the microorganisms required simple compound as food for their growth and then they can degrade the organic content present in the wastewater to release protons and electrons. So there was requirement of glucose-a simple sugar by the microorganisms for every 2 hours in cycle 3 and 3 hours in cycle 4. Whenever the voltage got decreased, there was glucose addition and was more than 2-4 times in both the cycles and as the reaction time increased, the glucose addition got reduced because organic matter in the wastewater might have got decreased indicating the treatment of wastewater. After 72^{nd}hr, maximum voltage and current was generated i.e., 0.120 V, 0.177 mA for DDMFC and 0.061 V, 0.088 mA for DMFC for cycle 3 and 0.202 V, 0.577 mA for DDMFC and 0.101 V, 0.286 mA for DMFC for cycle 4. By conducting three cycles with NSL industrial wastewater, it was concluded that voltage production increases with decrease in volume of the sample. So, furthermore experiments (cycles 5-8) were carried out with ICL industrial wastewater. In addition to distillery wastewater, sugar wastewater of the same industry was used instead of adding glucose and performance of the system was evaluated.

Voltage generation was more with ICL sample than with NSL sample because the concentration ofmicroorganisms was more in ICL sample. After 72^{nd}hr, maximum voltage and current was generated i.e., 1.252 V, 0.844 mA for DDMFC and 0.692 V, 0.452 mA for DMFC for cycle 5; 1.792 V, 1.47 mA for DDMFC and 0.918 V, 0.735 mA for DMFC for cycle 6; 1.578 V, 1.341 mA for DDMFC and 0.806 V, 0.672 mA for DMFC for cycle 7; 1.959 V, 1.496 mA for DDMFC and 0.976 V, 0.750 mA for DMFC for cycle 8.The volume of the sample was increased by 25 ml from cycle 5-7 and cycle 6 was to found be optimum for maximum voltage generation.

To check the performance of DDMFC with 125 ml of sample (optimum dosage), cycle 8 was carried out without the addition of sugar wastewater and glucose. Maximum voltage of 1.959 V and current of 1.496 mA after 240^{th} hour was generated in cycle 8, because sugar wastewater may be toxic for the microorganism present in the distillery wastewater. A longer reaction time was observed to reach the maximum voltage, because wastewater contained more of polysaccharides.

Table 4: Maximum voltage and current generated in DDMFC and DMFC

Cycle number	Time (hour)	Maximum Voltage (V)		Maximum Current (mA)		Power * (mW)	
		DDMFC	DMFC	DDMFC	DMFC	DDMFC	DMFC
3	79	0.120	0.061	0.177	0.088	0.021	$5.368x10^{-3}$
4	79	0.202	0.101	0.577	0.286	0.117	0.029
5	79	1.252	0.692	0.844	0.423	1.056	0.293
6	79	1.792	0.918	1.470	0.735	2.646	0.666
7	103	1.599	0.806	1.341	0.672	2.144	0.541
8	247	1.959	0.976	1.496	0.750	2.931	0.732
	655	2.012	1.007	1.873	0.937	3.768	0.944

3.3 Correlation of COD Removal on Maximum Voltage

A good positive correlation between maximum voltages and percentage of COD removals was found from four experiments operated. As demonstrated in figure, the correlation coefficient (R=0.939) of these two

parameters was acceptable. This particularly means the apparatus in addition can be used as a COD monitoring system to predict the COD reduction over the period of digestion from the increase of maximum cell voltage of the MFC. As a result, electrogenic digestion generated substantial number of electrons which were transferred to aerobic compartment to react with oxygen from air pumping system. As the organic matter got degraded, percentage of COD removal got increased and hence the voltage got decreased.

3.4 COD Removal Efficiency

COD removal efficiency was observed in DDMFC. Distillery wastewater showed its potential for COD removal indicating the function of microbes, present in wastewaters in metabolizing the carbon source as electron donors. It is evident from experimental data that current generation and COD removal showed relative compatibility. COD removal efficiency increased from 22% to 93% from first day to last day (7days/cycle) in all the cycles. The COD removal per day for all cycles is given table 4.4 and 4.5. In spite of addition of glucose, there was no increase in COD value because immediately after the addition of glucose it was consumed by the microorganisms and also 3g of glucose contains 0.12 g of COD, it is very less. There was also a considerable change in the colour of the wastewater.

Table 5: COD removal in each cycle

Time (hour)	COD (mg/L)				
	Cycle 3	Cycle 4	Cycle 5	Cycle 6	Cycle 7
0	36280	32000	22525	25686	35600
31	28382	25012	16000	18542	25800
55	21600	19122	11450	122781	18000
79	16900	14000	7898	8500	15000
103	12500	10722	4000	4800	12000
127	9320	7625	3020	3154	6200
151	8152	6417	-	-	4900

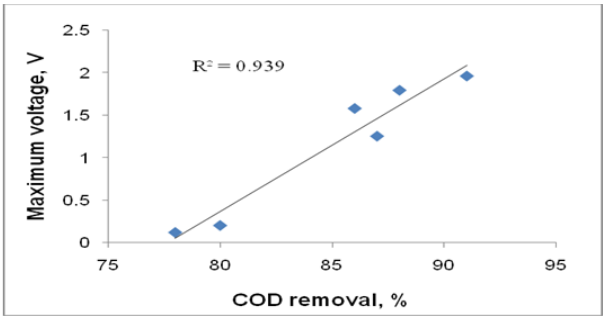

Figure 3: Correlation between maximum voltage and percentage of COD removal for all the cycles

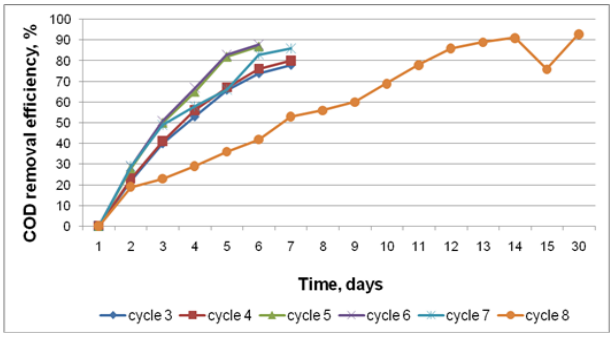

Figure 4: COD removal efficiency

IV CONCLUSION

Utilizing wastewater (renewable energy) for the production of renewable energy (bioelectricity) from anaerobic treatment is a feasible, economical and sustainable alternative. To minimize costs and to make a more durable cell, precious metals and costly membranes were avoided. Performance and stabilization tendency with respect to power generation was found to be dependent on the volume of the substrate and 125 ml of wastewater was found to be optimum. A maximum voltage and current of 1.959 V, 1.496 mA for DDMFC and 0.976 V, 0.750 mA for DMFC were generated for 125 ml of distillery wastewater (cycle 8). For re-stabilization of the system (cycle 8), sugar wastewater and glucose were added and sugar wastewater was found best for re-stabilization. After re-stabilization, a maximum voltage and current of 2.012 V, 1.873 mA for DDMFC and 1.007 V, 0.937 mA for DMFC for (cycle 8). The observations indicate that the MFC technique can be a potential method for disposal of distillery waste in view of the environmental protection. Up to 93% and 92% of COD and BOD removal efficiency respectively were achieved with DDMFC. A good positive correlation between maximum voltages and percentage of COD removals was found from four experiments operated. Major advantages of energy produced from wastewater are the absence of environmental emissions, simultaneous recovery of energy and wastewater treatment. Connecting MFCs in series generates more electricity than with a single MFC. A good understanding of the acclimatization of the communities in MFCs and their response to environmental perturbations would reduce the perceived risks and accelerate the adoption of MFCs.

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