Performance of two-chambered Microbial Fuel Cell (MFC) at different pH anode microenvironment using Palm Oil Mill Effluent (POME) as substrate

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Abstract. Microbial fuel cell (MFC) represents a new method for producing electricity from the oxidation of organic matter. In addition, MFC offers an effective wastewater treatment. The feasibility of using POME wastewater as a substrate was investigated through a two-chambered MFC operated in batch mode for 12 days. The performance of MFC was evaluated under three different anode pH microenvironments of acidic (pH 4), neutral (pH 7) and alkaline (pH 8). Results of experiments indicated that the MFC reactor was able to generate electricity and treat POME wastewater that acted as substrate for MFC. The performance of MFC was found to be dependent on the anode pH microenvironments. Higher power density was observed at neutral condition compared to acidic and alkaline conditions. Furthermore, significant reductions in chemical oxygen demand (COD) in anode chambers were found due to the changes of pH in anode microenvironment. This indicated that effective wastewater treatment of POME in MFC batch experiments. In conclusion, MFC provides an alternative, sustainability and effective method to generate electricity and effectively treat wastewater.

1. Introduction

Microbial fuel cell (MFC) is a biochemical reactor which has the ability to generate electricity and treat wastewater simultaneously. MFC generates electricity from the redox reaction with the aid of a catalytic reaction of microorganisms [1].

The catalytic reaction is initiated by the metabolic activity from the oxidation of organic matter by microorganisms in the anode chamber [2]. This reaction generates electrons (e^-) and proton (H^+) ions. Electrons travel to the cathode chamber through an external circuit under an external resistance while proton ions diffuse to the cathode chamber through cation exchange membrane (CEM). At the cathode chamber, electrons reduce oxygen which acts as the electron acceptor to produce water. The process in the MFC is advantageous as it does not require the thermodynamic conversion step [3,4]. In addition, the biological process offers an economical method for both electricity generation and wastewater treatment.

MFC is classified as an alternative sustainable technology as it adopts the 'Waste to Wealth' concept where this technology uses biomass waste to act as a carbon source to fuel the cell. Patil *et al.* [5] have reviewed on variety of wastewater that has been used in MFC. To make MFC competitive with other technologies, agriculture and industrial wastewater are being considered to be the most suitable candidate as it is a cost free waste and sustainable resource [6]. Renewable energy generated by MFC, which uses different types of wastewater as carbon source, is relatively new in Malaysia.

Palm oil is an important economic product in Southeast Asia as well as in Malaysia. The palm oil mill is the biggest sector in manufacturing sector in Malaysia, thus establishing Malaysia as the world largest producer of palm oil. In 2008, it accounted for approximately 47% of world palm oil production and 54% of world exports [7, 8]. The palm oil sector produces abundant wastewater known as Palm Oil Mill effluent (POME). POME is a viscous brown liquid containing high chemical oxygen demand (COD) with fine suspended solid (SS) [9]. POME must be treated to

meet discharge regulations. Currently, POME is no longer seen as a waste, but instead a valuable energy source through MFC approach of treating wastewater organic matter and thus generates useful amount of renewable energy.

Generally, in a two-chambered MFC reactor, wastewater with high organic matter is placed in the anode chamber. The typical catalytic reaction is initiated by the bacteria in the anode chamber. The anode pH microenvironment is an important factor as bacteria activities depend on the pH conditions. Therefore, anode pH microenvironment is influenced by the bacteria activities and affected the e^- and H^+ generation mechanism.

This paper focuses on electricity generation and wastewater treatment using a two-chambered MFC utilizing POME wastewater as substrate with different anode microenvironments. These investigations not only lead to MFC approach for alternative renewable energy and wastewater treatment but also contribute to the sustainable practice and waste to wealth concept.

2. Methods

2.1 Laboratory scale Microbial Fuel Cell (MFC) construction

Three identical two-chambered MFC reactors were setup in this study. The MFC reactors were composed of two 500 ml volume conical flask using salt bridge as a separator or act as a CEM. Salt bridges of 15 cm length and internal diameter of 0.8 cm were prepared using a mixture of KCl (Merck, Germany) and agar (Merck, Germany) in ratio of 1:1. The stoppers were equipped for sampling ports, wire input, salt bridge, and gas inlet and gas outlet. The electrodes used were carbon rod with projected area of 11.46 cm^2 . Prior to use, electrodes were soaked overnight in deionized water to remove organic contamination on the electrode surface. Electrodes were connected with copper wire through the resistance $3.3 \text{ k}\Omega$. Copper wire was sealed with rapid epoxy sealant to prevent reactions between copper wire and wastewater sample which might lead to corrosion. A multimeter (Fluke 289 True RMS Multimeter, Fluke Corporation, USA) was used in the setup to measure the produced voltage.

2.2 Wastewater composition

POME samples were collected from the Bukit Kerayong palm oil mill in Selangor, Malaysia. POME was used as a substrate for electricity generation in the anode chamber of the fuel cell. The average characteristics of POME wastewater samples used were pH 4.59, conductivity $9.39 \text{ }\mu\text{S/cm}$, suspended solid (SS) $87\,566 \text{ mg/L}$ and chemical oxygen demand (COD) $98\,000 \text{ mg/L}$. POME is considered as complex substrate in nature due to its composite nature and high-biodegradability. ($\text{BOD/COD} \sim 0.5$).

Domestic wastewater (DWW) samples were collected from a local Sewage Treatment Plant in Universiti Teknologi MARA, Selangor, Malaysia. DWW was used as a biocatholyte which also acts as a cathode electrolyte in the cathode chamber of the fuel cell. The characteristics of DWW were pH 6.78, conductivity $246.86 \text{ }\mu\text{S/cm}$, SS 98.87 mg/L and COD 264 mg/L .

2.3 Microbial Fuel Cell (MFC) operation

Each assembled MFC reactor setup in Section 2.1 was filled with 500 ml of aerated domestic wastewater (DWW) as biocatholyte that acts as a cathode electrolyte in all experiments. Air was supplied in the cathode chamber to promote aerobic condition.

On the other hand, the anode chamber was filled with 500ml of diluted POME at different pH as substrate. Anode pH of POME was adjusted to pH 4, pH 7 and pH 8 respectively. The pH adjustments were conducted by using weak acid or base solutions to the design anode pH values. At the anode chamber, nitrogen (N_2) gas was purged within the POME sample for at least ten minutes to ensure the anode chamber was free from oxygen and hence created an anaerobic condition. Finally, adhesion promoter seal was applied at anode chamber to ensure the anaerobic condition.

Experimentst were operated at regular room temperature ($28 \pm 2^\circ\text{C}$) in a batch mode as suggested by Du *et al.* [10]. The amount of voltage produced during the experimental work was measured using a digital multimeter for each assembled MFC reactor. Data was recorded hourly for

12 days. Experiments were carried out at different anode pH to evaluate the effects of pH on the electricity generation and wastewater treatment.

2.4 Electrochemical analyses and calculations

The performances of MFC were studied in terms of electricity generation at different anode pH microenvironments of pH 4, pH 7 and pH 8. Electricity generation evaluations include electrochemical analysis on the effects of anode pH microenvironment and polarization behavior.

2.4.1 Electrochemical analysis on effects of anode pH microenvironment

The recorded data from section 2.3 was evaluated. Graph according to anode pH microenvironments of current versus time and power versus time were plotted and discussed respectively.

2.4.2 Electrochemical analysis on polarization behavior

The performance of MFC was investigated based on voltage output of fuel cell after a steady state. The steady state occurs when the voltage reading become stable after operation. The voltage and current were measured using a digital multimeter and was monitored daily for a period of 12 days. Voltage and current were converted to power according to Equation (I).

$$P = iv \quad \text{Equation (I)}$$

where P is power in Watt (W), i is current in Ampere (A) and v is voltage in Volt (V). Power density and current density were calculated using the normalized surface area of the anode electrode. Power curves were derived from the polar curves [11]. Power curves indicated the maximum power density produced from the MFC systems. Polarization curves were formed by varying the external resistance from 100 Ω to 100 k Ω in parallel circuit connection and were plotted as current density against voltage output. Internal resistances of the MFC systems were determined using the slope of linear regression line of the polar curves [12].

2.5 Wastewater analysis

Wastewater parameters were determined using standard method APHA *et al.* [13]. The POME samples were extracted on day 1 and 12 to determine the efficiency of wastewater treatment in anode chamber. COD was determined. Samples were analyzed in triplicates until standard deviation less than 5% was achieved.

3. Results and discussion

3.1 Electrochemical analysis

3.1.1 Effects of pH on electricity generation

The performance of two-chambered MFC in electricity generation was influenced by the variation of anode pH microenvironments. Figure 1 shows current production utilizing POME as anode substrate at different anode pH microenvironments. In addition, Table 1 shows the MFC's performance at different anode pH microenvironments. Generally, significant variation in current production and electrogenic activity over time was observed from the output trend. The trend can be divided into three phases, namely, ascending phase, stationary phase and declining phase. This observation is consistent with Min *et al.* [14] and Kaewkannetra *et al.* [15].

Figure 1: Current production from MFC reactor at different anode pH microenvironments

Table 1: MFC performance at different anode pH microenvironments

Performance	Acidophilic [pH 4]	Neutral [pH 7]	Alkaline [pH 8]
Maximum voltage [mV]	25	35.6	4.31
Maximum power [mW]	0.19	0.38	0.006
Maximum current [mA]	0.007	0.01	0.001

Significant variations in current production and electrogenic activity over time were observed from the output trends shown in Figure 1. The anode with microenvironment pH 7 recorded the optimum anode pH microenvironment in a two-chambered MFC. At anode pH 7, the voltage of 31 mV was observed during the initial phase of operation. A steady increase in voltage output was observed and finally approached a maximum of 35.6 mV at 190 h. In addition, maximum current of 0.01mA at 3.3 k Ω external resistance was observed. At the optimum anode microenvironment pH of 7, the electrogenic bacterial growth is favorable which promotes the growth of microorganisms for electricity generation and contributed to the effective extracellular electron transfer at this anode pH microenvironment [16]. Neutral anode at pH 7 microenvironment generated higher power compared to acidic and alkaline conditions. This is closely related with the active degradation carried out by the bacteria and internal resistance of MFC system.

At acidic anode pH of pH 4, the voltage of 22 mV was observed and later approached maximum of 25 mV at 200 h. In addition, maximum current and maximum power was 0.007 mA and 0.19 mW at 3.3 k Ω external resistance respectively. At anode pH of 4, it promoted the growth of acidogenic bacteria. The metabolic activity of acidogenic bacteria caused accumulation of methanol as by-product hence decreased the performance. This created a conductive environment for acidogenic bacteria which had a lower pH around pH 6. This findings is supported by Mohan *et al.* [17] and Ishii *et al.* [18].

Alkaline condition at pH 8 recorded slow increased in both current and power generation and reached its maximum of 0.006 mA and 0.01 mW at external resistance 3.3 k Ω . The slow increase production of current and power was due to low activity of the microorganism in POME under alkaline condition and thus resulting in low current and power generation.

In this study, anode microenvironment of pH 4 and pH 7 involved all three phases. However, at pH 8 only two phases was observed, namely, ascending phase and stationary phase. This was due to the low electrogenic activity as the microenvironment was not favorable for microorganisms. A sudden rise in current of 0.01 mA and 0.007 mA and power of 0.33 mW and 0.15 mW were observed at anode pH 7 and pH 4 respectively. These immediate fluctuations might be due to the difference potential between the two electrodes. In addition, declining phase were to be expected as the organic matter in POME and DWW samples were not replaced in a batch mode experiment and thus became the limiting factor that caused current and power reduction. It was observed that the ascending sequence performance was at pH 8, followed by pH 4 and reached the best performance at pH 7. A consistence increase in MFC performance was observed might be attributed to the adaptation tendency to the anode pH microenvironments.

Generally, in the anode chamber, microorganisms activities of both proton generation and consumption occur at the same time, but a balance need to be established between proton generation and consumption. This can be achieved by controlling the pH of substrate used. The neutral anode microenvironment of pH 7 showed the highest performance in power and current. This was also reported by Jadhav and Ghangrekar [19] where the amount of proton produced in the anode chamber should penetrate through cation exchange membrane (CEM) and consumed at cathode at the same rate for cathodic reaction. However, this condition did not occur at pH 4 and pH 8 anode microenvironment. Low values of current, power, power density and current density at pH 4 and pH 8 in the anode microenvironment may be due to slow production rate of proton and electron in the anode chamber and poor proton transfer across CEM which at this pH is favorable for methanogenesis.

Some differences and similarities were found when comparison was made against previous studies. Behera *et al.* [20] reported that power generation decreased with decreased in influent pH. However, this study observed an optimum output at anode pH neutral of pH 7 followed by acidic of pH 4 and later alkaline of pH 8. Behera *et al.* [20] and Behera and Ghangrekar [21] reported an optimum anode pH at pH 8 where the MFC reactor generated 304 mV and 680 mV respectively. These showed a big difference of 71 to 157 times the output of present study. This high result is closely related to MFC setup and materials, namely, single chamber and platinum [21] and dual-chamber and Nafion 117 [20]. Besides the studies discussed, other researcher have also found that at acidic condition can also resulted as an optimum condition for maximum output compare to neutral and alkaline condition[3,22]. Raghavulu *et al.* [3] and Raghavulu *et al.* [22] observed maximum voltage of 324 mV and 632 mV and maximum current of 1.6 mA and 5.18 mA as the metabolic activity of acidogenic bacteria reacts well in this condition. On the other hand, there are other studies agreements on the optimum pH at neutral pH of 7. This condition is suitable for electrogenic bacterial growth and assist in the electron production in anode chamber [23,21]. Therefore, selection on appropriate anode pH microenvironment is important which triggers the biological pathway and electron production phenomena in the MFC reactor.

3.1.2 Polarization behavior

Polarization behavior of MFC during operation was recorded at various external resistances (100Ω-100kΩ) to study the electron discharge at various anode pH microenvironments. Polar curves helps to determine the fuel cell behavior at various external resistances. Figure 2 shows a polar curve at various anode pH microenvironments while Table 2 shows the performance at different anode pH microenvironments of MFC reactor. It was observed that voltage generation decreased with increased in resistance which indicated a typical fuel cell behavior. A consistent decreased in current also was observed with increased in resistance. Increased in resistance caused reduction in electron discharge while low resistance increased electron discharge and thus resulting high voltage reading at all different anode pH.

Figure 2: Polar curve at various anode pH microenvironments

Table 2: MFC highlighted performance at different anode pH microenvironments

Performance	Acidic (pH 4)	Neutral (pH 7)	Alkaline (pH 8)
Maximum power density (mW/m ²)	2.86	3.21	1.15
Maximum current density (mA/m ²)	0.046	0.049	0.034
Internal resistance (kΩ)	5.5	5.3	6.1

Maximum power density showed highest electron discharge and also indicated the low internal resistance of the MFC system when power density is at its maximum performance. The observed maximum power density at acidic, neutral and alkaline were 2.86 mW/m² at 5.5 kΩ, 3.21 mW/m² at 5.3 kΩ and 1.15 mW/m² at 6.1 kΩ respectively. Alkaline and acidic condition showed maximum power density at comparatively low performance as the internal resistance at alkaline and acidic condition was very high (5.5-6.1 kΩ). Meanwhile, the maximum power density had low internal resistance of 5.3 kΩ which generated more power due to enhance in higher electron discharge. This enumerates the feasibility of neutral condition in effectively discharging the electrons compared to acidic conditions. In addition, at lower resistance condition, the electrons move more easily through the circuit compared to at higher resistance. Higher oxidation reaction by the microbes is expected at a lower resistance and therefore the MFC can be operated at lower resistance to remove the organic matter at a higher rate (Jang *et al.*, 2004). It was observed that a rapid voltage drop at lower external resistance and voltage was getting stabilized at higher external resistance. The rapid voltage drop and slow stabilization of voltage were due to the rate of electron discharge at lower resistance.

In comparison with other anode pH microenvironment, acidic condition showed a stable voltage output until 300 h. This suggested that the operation of MFC must be controlled consistently at optimum anode pH 7 throughout the experiment. Appropriate selection of anode pH microenvironment will support the continuous electron discharge from substrate degradation [22] as the anode chamber controls the kinetics of e⁻ transfer from microorganism to the anode [22].

Raghavulu *et al.* [3] and Raghavulu *et al.* [22] reported that the optimum pH was slightly acidic condition of pH 6 using chemical wastewater as substrate. A power density of 12.72 mW/m² and 62.7 mW/m² were reported using single chamber and two-chamber respectively [3,22]. The high power density observed was due different setups. Studies carried out by Ghangrekar with fellow researcher, found that two-chambered MFC can produces as high as 600 mW/m² at alkaline pH of pH 8 however this was due the suspension sludge concentration and substrate were oxidized by other anaerobic microorganism in the anode chamber [20,21]. Nevertheless, this study is in-line with Jadhav and Ghangrekar [19] which reported that the pH near neutral is the optimum pH in the anode chamber. Jadhav and Ghangrekar [19] observed a power density of 17.1 mW/m² and 15.2 mW/m² with internal resistance of 523 Ω and 547 Ω at anode pH of 6.5 and 7 respectively. This was due the pH condition near neutral was ideal for the bacteria activities to oxidize and reduce proton and electron in both chambers hence high output with low internal resistance was observed.

3.2 Wastewater treatment

Besides power generation, wastewater treatment in the anode chamber was also observed during MFC operation. The COD reduction obtained were 65 %, 55 % and 43 % at acidic, neutral and alkaline anode pH condition respectively. In contrary to the power generation, acidic anode pH 4 microenvironment showed highest percentage COD removal followed by neutral microenvironment at anode pH 7 and finally alkaline at anode pH 8. The experimental results clearly demonstrated that the MFC performance is dependent on the anode pH microenvironments. The increase of COD removal efficiency was observed when pH of anode microenvironment decreased. This is attributed to the shorter acclimatization period of the anaerobic bacteria inside the MFC as acidic condition represents the natural POME environment at the initial stage of MFC operation.

Besides, acidic microenvironment in anode chamber may be attributed to the efficiency of methanogenic bacteria in metabolizing the substrates [22] and supported the growth of

methanogenic bacteria and other non-specific microbial populations. Moreover, acidic condition generally enhance the function of methanogenic bacteria and creates conducive environment for the proliferation of acidogenic bacteria, where complete reduction of substrate was feasible [22]. Methanogenic bacteria functioned effectively in near pH 7 while acidogenic bacteria had lower pH optimum around pH 6 and were insensitive to acidic conditions [24].

Previous studies on increase of anode pH reported an improved in percentage of removal COD ranged 55.76%-85.4%, 58.98%-88.5%, 47.8-92.6% at pH 6, 7 and 8 respectively [3, 20, 22]. The improved COD removal was due to adaptation of bacteria to the initial condition which is electrochemically active and later favors the condition near neutral which promote bacterial growth [19].

3.3 POME as substrate

The high levels of COD and conductivity of POME encourage the electricity generation. The importance of high conductivity for maximizing electricity generation has been highlighted by Huang and Logan [25]. They reported that an increased in the power density of 245% when the solution conductivity was raised from 0.8 $\mu\text{S}/\text{cm}$ to 10.2 $\mu\text{S}/\text{cm}$. As the conductivity of solution increased, the internal resistance for power generation in the system became lower and facilitated the transport of ions through the anode biofilm [21]. This condition might be due to the presence of electrochemically active microorganisms. The solution conductivity of POME sample used in this study was about 9.36 $\mu\text{S}/\text{cm}$ which was considered conducive. Therefore, the POME sample is a highly potential candidate for MFC substrate for electricity generation as it is conducive and is readily biodegradable waste with high COD.

POME samples used in this study was acidic in nature and neutralization of the wastewater is necessary for biological treatment and to promote growth of bacteria in the wastewater. Higher acidity or alkalinity of wastewater affects both wastewater treatment efficiency and the microenvironment of the reactor. The neutral pH condition needs to be maintained in order to achieve favorable biological treatment condition for microorganism growth and metabolic activities hence maximizes electricity generation.

4. Conclusions

This study demonstrated electricity generation and effective wastewater treatment utilizing POME as substrate in a two-chambered MFC reactor. This study revealed that the performance of MFC was found to be dependent on the anode pH microenvironments. It was demonstrated that anode pH microenvironments affects the anodic bacterial activity and the overall performance of the MFC. From the experiments it was found that the bacteria in POME can tolerate anode pH at acidic as low as pH 4 and at alkaline pH as high as pH 8. The optimum condition at neutral pH 7 anode microenvironment was observed with the performance on electricity generation of 35.6 mV, 0.38 mW and 0.01 mA. This condition was favorable for higher power generation and high treatment efficiency compared to acidic and alkaline microenvironment conditions. In addition, 55 % COD removal efficiency at pH 7 produce maximum current. The COD removal efficiency indicated the functioning of MFC as an alternative wastewater treatment in addition to renewable energy generation.

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