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The behaviour of membrane less sediment microbial fuel cell in the terms of bioremediation and power generation

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ABSTRACT

Aims: To study the performance of SMFC in the terms of power generation and toxic metals removal. This study was also focused on the characterization of SMFC electro-microbiology.
Methodology and results: A SMFC was designed and loaded with sediment and overlying water. This SMFC was synchronized with wireless data logger acquisition system. The toxic metals removal capacity was measured by atomic absorption spectroscopy. The characterization of SMFC bacteria was done by 16S rRNA. In this study the experiments were carried out in a dual-chamber SMFC with external resistances 30 kΩ-50 Ω. The SMFC was produced power about 630 mV with maximum power density 40 mW/m^2 and current density 250 mA/m^2. After 120 days of operation, SMFC removed cadmium and copper about 22.6 and 150 mg/kg, respectively. The SMFC also showed high cadmium (86%) and copper (90%) removal at pH 7.0 and temperature 40 °C. The most dominant bacterial community at anode and cathode was identified as *Pseudomonas* spp. which could be function as exoelectrogen.
Conclusion, significance and impact of the study: The results indicated that the SMFC system could be applied as a long term and effective tool for the removal of cadmium and copper contaminated sediments and supply power for commercial devices. The *Pseudomonas* spp. may be used as a genetic donor for the other non-exoelectrogens strains.

Keywords: Electrodes, external resistance, power generation, sediment microbial fuel cell, toxic metals

INTRODUCTION

Sediments are important components of aquatic environment. The quality of water depends on the exchange of soil and water substrates and marine bottom. The surface payer of sediments consists significant quantity of toxic metals and organic matter, thus probably impending integrity of the ecosystem (Schievano *et al.*, 2017). The sediment oxidized layer surface inhibits diffusion of most toxic metals into the marine water. The industries like electronic and electroplating are causing a major issue of pollution by generating wastewater containing contaminants like toxic metals pose a serious danger to animals, humans and the environment (Abbas *et al.*, 2015; Xia *et al.*, 2015). Therefore, it is compulsory to treat industrial wastewater consisting toxic metals before to its discharge.

Several conventional metals removal approaches like membrane filtration, ion-exchange, coagulation-flocculation and chemical precipitation are used. They are useful but they confront some major drawbacks like extra chemical consumption, high energy demand and production of a huge amount of toxic waste sludge (Abbas *et al.*, 2014a; Hsu *et al.*, 2017). Microbial fuel cells (MFCs) systems have recently been of great concern as a most rising technique to treat industrial wastewater with power production. The sediment microbial fuel cells (SMFCs) or benthic microbial fuel cells (BMFCs) are an appropriate employment of MFC to remediate industrial wastewater and generate electricity from the electro-potential difference between anoxic sediments and oxic water present in the water body (Xu *et al.*, 2017). There general model of SMFCs contains of anode buried in marine sediment and the cathode is positioned in surface water. Microorganisms in the sediment and wastewater metabolize organic compounds, producing protons and electrons. Electrons are shifted from cathode to anode via an external circuit and combine with oxygen on cathode to generate water (Reimers *et al.*, 2001; Abbas *et al.*, 2017a). In this study the capacity of SMFCs to generate power and toxic metals removal was evaluated.

MATERIAL AND METHODS

Sediment sampling

The marine water and sediment (0-20 cm depth) were sampled from the Bayan Lepas (Pulau Pinang, Malaysia) industrial zone. The sediment was collected by using a ponar type sampler (2.4 L, 86475 Gene Lassere BLVD, USA). They were placed into clean polycarbonates jars

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(Kimax Kimble, Fisher brand) and shifted to the laboratory.

SMFC construction and operation

Three replicas of SMFCs were designed from plexiglass. Firstly, 767 g of wet sediments was supplemented to SMFC and then overlaid with 200 mL of the sampled marine water. For each SMFC, fifteen connected pieces of graphite plate (4 cm x 2 cm) were vertically inserted into the sediment with 1 cm gap as an anode array. In the surface water another graphite plate with same dimension were planted about 4 cm above the anode. The rubber sealed copper wire was used to connect anode and cathode terminals and silicone rubber was also used to cover the bonding sites prevent from leaking and corrosion. The overall SMFC dimensions were 17.6 cm x 8.7 cm (length × width) as shown in Figure 1. The lengths of cathode and anode chambers were 9.6 cm and 8.0 cm, respectively. Aeration was facilitated near cathode via commercially available air pump (JAD aquarium, electrical product Co. Ltd.) at a depth of 8 cm from the top marine water level in all these SMFCs. The SMFCs was connected with wireless data logger which attached with PC.

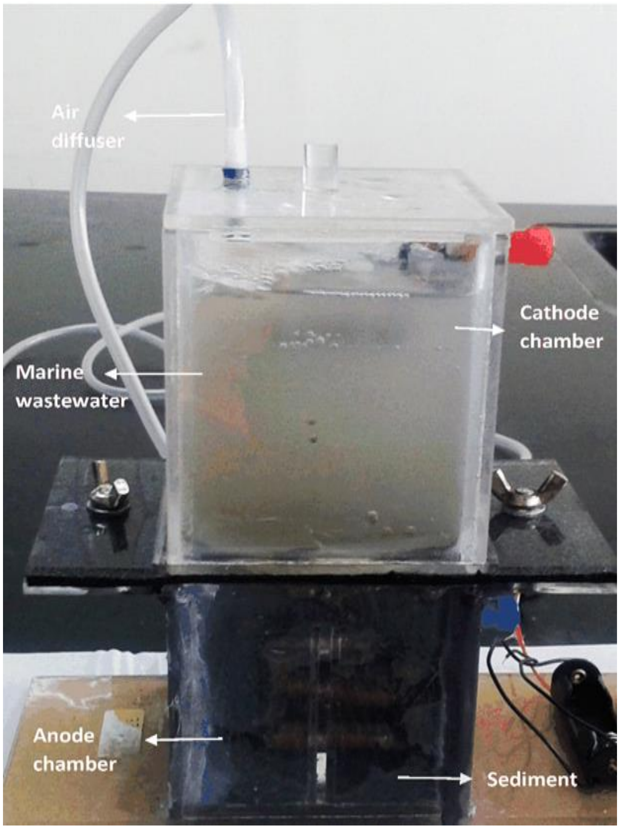

Figure 1: Prototype of SMFC.

Current/power

The ohm's law (R=V/I) was used to measure the current, where V is voltage and R is the external resistance (Ω). Power was calculated as P=VI. The various external resistances were used to measure the maximum power density with 15 min interval time for allowing the voltage to be stabilized. The slope of polarization curve was made to calculate the external and internal resistance for optimum performance of SMFC.

Toxic metals removal rate

The amount of cadmium and copper in the raw sediment samples were analyzed by sequential extraction method and compared with the sediment guidelines of Hong Kong (Chapman *et al.,* 1999). After operation of SMFCs, the concentrations of cadmium and copper were measured by atomic absorption spectroscopy (A3G graphite furnace atomic absorption spectrometer, USA) and compared with the initial concentrations. The cadmium and copper removal efficiency was also analyzed at different pH (1.0-13.0 and temperatures (0-80 °C) compared with power density generation.

Microbial characterization

After 120 days incubation, electrodes were dislodged surface marine water and sediment. Then surface of electrodes was washed with a stream of sterile freshwater to get rid of visible debris. A sterile razor blade was used to scrape vigorously about 1 mm biomass on the graphite electrodes into 1.5 mL Tris-EDTA (TE) buffer. DNA was isolated using commercial UltraClean DNA Isolation (Carlsbad, CA, USA). The universal primers Eubac27F (5'-AGAGTTTGATCCTGGCTC AG-3') and 1492R (5'-GGTTACCTTGTTAC GACTT-3') were used to amplify the bacterial 16S rRNA gene. The obtained genes were analyzed by NCBI blast.

RESULTS AND DISCUSSION

Physio-chemical properties of sediment

The physical and chemical properties of sediments are provided in Table 1.

Table 1: Physiochemical properties of sediment collected from Bayan Lepas, marine stream.

Parameters	Untreated sediments
Color	Dark brown
Temperature (°C)	25-35
pH	6.0-8.0
Sand (%, w/w)	17.9 ± 5.8
Silt (%, w/w)	85.4 ± 72.5
Clay (%, w/w)	4.5 ± 2.3
Carbon contents (%, w/w)	3.0 ± 1.3
Water contents (%, w/w)	50.5 ± 3.70
Electrical conductivity (μS cm^{-1})	498 ± 6.11

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Voltage generation

The voltage of SMFC was monitored until 120 days as shown in the Figure 2. The voltage was increased in the initial 60 days and gained the maximum value of 630 mV. The high redox potential was most likely the cause of such sharp increase of voltage. The high voltage production by SMFC was directly proportional to concentration of biodegradable organic matter. The high voltage production also indicated that the electrodes of SMFC were populated by exoelectrogens for electrons transfer and the current generation in SMFC (Zhou *et al.*, 2016).

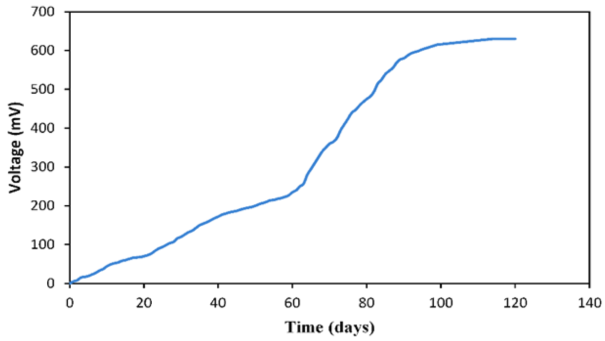

Figure 2: Voltage measurement of SMFC.

Polarization curve

The polarization curve was plotted against the power density and current density at different resistances (50 Ω-30 kΩ) to understand the SMFC behavior as shown in Figure 3. At 50 Ω external resistance the highest power density (40 mW/m^2) was achieved. This phenomenon was interpreted by the fact that the slowly and easily biodegradable sediment organic matters were oxidized by SMFC exoelectrogens (Hong *et al.*, 2009). The lowest power density (7 mW/m^2) was achieved under 30 kΩ external resistance and highest was obtained under 50 Ω. This proved that the SMFC performance was affected by external resistance because the activity of exoelectrogenic community depends on external resistance.

The external resistance could affect the availability of anode for exoelectrogenic bacterial activities result in influence the anode potential and biofilm formation on electrodes. As a result, under different external resistances the internal resistances were different from each other. The SMFC at 50 Ω had a smallest internal resistance of 20 Ω. The researches in the previous studies mainly focused on the enhanced of SMFC power generation by rotating of cathode for the availability of oxygen and used the cellulose and chitin as a biodegradable substrate (Logan, 2015). However, few studies have concentrated on the simultaneous power generation and bioremediation could be increased by improving SMFC configuration. This study also focused the power generation and in-situ remediation by improving the SMFC configuration.

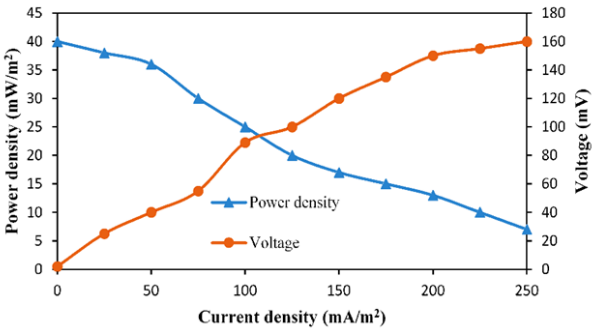

Figure 3: Polarization curve of SMFC.

Toxic metal removal

The exoelectrogenic bacteria were removed about cadmium (22.6 mg/kg) and copper (150 mg/kg) after 120 days of SMFC operation. The highest heavy metals removal rate until 60 days and then reduced steadily as shown in Table 2. The removal rate in the first 20 days was very slow because the bacteria need time to adjust with the medium environment. The reduction of cadmium and copper entirely depends on the oxidation of acetate into the anode chamber to transfer the electron these bacteria and reduction of cadmium and copper proceeded rapidly in the cathode chamber. Other organic compounds also reduced by these exoelectrogenic bacteria. However, some few studies were carried out on the reduction of many toxic metals by many bacteria. *Geobacter sulfrreducens* and *Geobacter metallireducens* were reduced the soluble uranium (VI) and chromium (VI) into insoluble and less toxic forms uranium (IV) and chromium (III), respectively. The *Geobacter lovleyi* reduced tetrachloroethylene and trichloroethylene to *cis*-dichloroethylene. The *methanobacterium palustre* was able to reduce the dehalogenated 2- chlorophenol to phenol (Abbas *et al.*, 2017 b, c).

Table 2: Performance of SMFC for the removal of cadmium and copper compared with sediment quality guidelines Hong Kong (SQGHK).

Toxic metals	Days of SMFC operation	Conc. (mg/kg)	SQGHK	
			LCEL[a] (mg/kg)	UCEL[b](mg/kg)
Cadmium	0	25.2	1.5	4
	20	18.6		
	40	15.1		
	60	9.7		
	80	7.5		
	100	5.4		
	120	3.4		
Copper	0	160.2	65	110
	20	147.4		
	40	125.8		
	60	70.6		
	80	40.8		
	100	30.7		
	120	10.2		

[a]Lower Chemical Exceedance Level
[b]Upper Chemical Exceedance Level

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Effect of pH on SMFC performance

To determine the effect of pH on the performance of SMFC, the power density and the removal of cadmium and copper were examined as shown in the Figure 4. The removal of cadmium and copper increases from 5% to 86% and 10% to 90% as pH raises from 2 to 7, respectively. Normally, bacteria require neutral pH to perform maximum. However, the bacterial pH also influenced by alkaline reactions of anode and cathode. Therefore, bacteria produce some weak acids to adjust with internal pH. The narrow range of pH 6-9 is needed for biofilm formation on electrodes. The lower pH near cathode associated with protons transfer to cathode and if protons not transfer to cathode the anode chamber pH becomes more acidic (Sajana *et al.*, 2013; Abbas *et al.*, 2014b). The Figure 4 also illustrates that pH also effects the power density of SMFC. The power density was increased 110.6 to 200.7 mW/m^2 as pH reached to 7. This showed that neutral pH played a positive impact on power generation. The sharp decrease of power density was pH 9 was possibly due to the diffusion of OH^- ion to anode chamber.

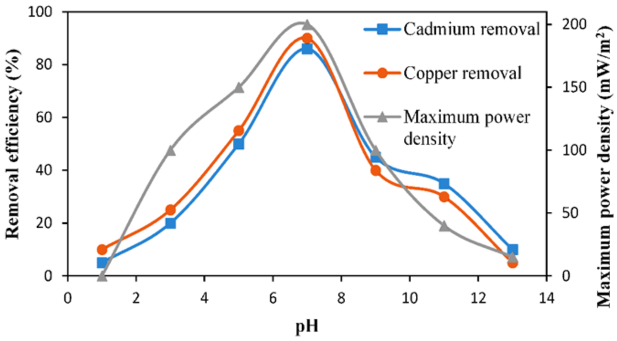

Figure 4: The effect of pH on the performance of SMFC.

Effect of temperature on the performance of SMFC

To see the effect of temperature on the performance of SMFC the graph was plotted against removal efficiency and temperature range (10-80 °C) as shown in the Figure 5. The cadmium and copper removal was increased from 5% to 82% and 8% to 88%, respectively as temperature raised from 10 °C to 40 °C. The power density was also maximum at 40 °C about 150.4 mW/m^2. The increased toxic metals removal and power generation may be due to the reduction of ohmic resistance and strengthen of metabolic pathways. The temperature has an inversely proportionally relation with ohmic resistance. The less biofilm developed at low temperature lead to reduced performance of SMFC. The higher temperature denatures the enzymes result in inactivation of metabolic activities. Different temperatures are needed for different exoelectrogenic bacteria for optimum metabolic activities (Abbas *et al.,* 2016).

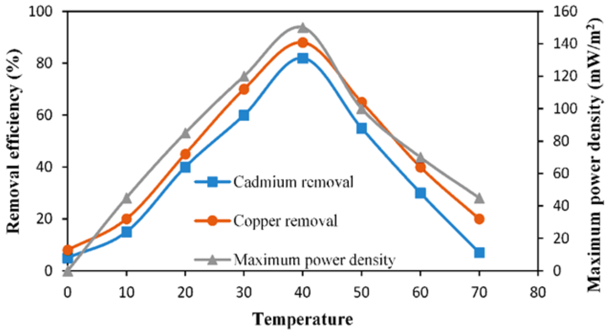

Figure 5: The effect of temperature on the performance of SMFC.

Exoelectrogenic community

The most dominant bacterial species on the both electrodes *Pseudomonas* strains as shown in the Table 3. These *Pseudomonas* strains were responsible for electrons transfer to the electrodes for power generation and remediation of toxic metals. Previous studies were mostly reported the *Geobacter* and *Shewanella* strains as exoelectrogens in the SMFCs but in this study the *Pseudomonas* strains were exhibited the SMFCs. Boon *et al.* (2008) also reported the *Pseudomonas* strains for power generation remediation of organic compounds.

Table 3: 16S rRNA gene sequence types obtained from clone libraries of SMFC electrodes.

Accession no. of 16S rRNA gene	Name of bacteria	Percentage homology
EU330371	*Pseudomonas* sp. BSi20432	100%
KR012218	*Pseudomonas* sp. JXH 225	100%
KR012205	*Pseudomonas* sp. JXH 211	100%
U22426	*Pseudomonas stutzeri* strain 19msn4	100%
JX177719	*Pseudomonas stutzeri* strain 2D54	100%

CONCLUSION

The SMFC power generation, effect of pH, temperature and external resistances were analyzed. The pH, temperature and external resistance affected the SMFC toxic metals removal efficiency and power density. The Power density was less at higher and lower pH and temperature. However, for full scale SMFC application, more researches are needed on other dimensions of SMFC like electrodes distance, surface area and nature of SMFC material. The energy production from SMFC is still low so there is need to more focus on which microbial community is potential candidate for exoelectrogens.

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