

Evaluation of microbial fuel cell (MFC) for bioelectricity generation and pollutants removal from sugar beet processing wastewater (SBPW)

Atikur Rahman, Md Saidul Borhan and Shafiqur Rahman

ABSTRACT

Bioelectricity generation from biodegradable compounds using microbial fuel cells (MFCs) offers an opportunity for simultaneous wastewater treatment. This study evaluated the synergy of electricity generation by the MFC while reducing pollutants from sugar beet processing wastewater (SBPW). A simple dual-chamber MFC was constructed with inexpensive materials without using catalysts. Raw SBPW was diluted to several concentrations (chemical oxygen demand (COD) of 505 to 5,750 mg L⁻¹) and fed as batch-mode into the MFC without further modification. A power density of 14.9 mW m⁻² as power output was observed at a COD concentration of 2,565 mg L⁻¹. Coulombic efficiency varied from 6.21% to 0.73%, indicating diffusion of oxygen through the cation exchange membrane and other methanogenesis and fermentation processes occurring in the anode chamber. In this study, >97% of the COD and up to 100% of the total suspended solids removals were observed from MFC-treated SBPW. Scanning electron microscopy of anode indicated that a diverse community of microbial consortia was active for electricity generation and wastewater treatment. This study demonstrated that SBPW can be used as a substrate in the MFC to generate electricity as well as to treat for pollutant removal.

Key words | bioelectricity generation, chemical oxygen demand, coulombic efficiency, microbial fuel cell, sugar beet processing wastewater, wastewater treatment

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INTRODUCTION

Over 18 million tons of sugar beet is produced annually in North Dakota and Minnesota, USA, to produce sugar from beets, and this is an important industry in the north central states (USDA 2014). For each ton of sugar produced, processing of beets requires 20 m³ of fresh water without considering reuse of it (Gutteck 1989). Combined with water contained in beets (about 75% of beet's weight), a large amount of wastewater is produced as a result of processing of beets. A part of this wastewater is reused in the processes; however, the remaining wastewater has to be treated in wastewater treatment facilities before discharging them into surface water. The sugar beet processing wastewater (SBPW) is enriched with high organic loadings, and treatment of the wastewater requires a large amount of energy. Often, untreated wastewater is discharged into nearby streams, which may cause water pollution including odor and bad taste, and impacting city water supply and aquatic life (Storry 1996).

Sugar beet processors use water for three main purposes: cooling circuits, wash and transport circuit, and processes (Iza *et al.* 1990). Wastewater released from the washing and transport is low in pollutant loadings, but the wastewater produced in different components of the process phase is high in pollutant contents including organic loadings and Steffen waste (Nemerow 1978). The high organic loading in the process wastewater is mostly due to the presence of sugars and organic materials transported with beets, which results in high biochemical oxygen demand (BOD) and chemical oxygen demand (COD) concentrations. In untreated effluents of SBPW, the BOD₅ concentration may range from 4,000 to 7,000 mg L⁻¹, while COD concentration may be in the order of 10,000 mg L⁻¹ (Güven *et al.* 2009) or even more. Wastewater resulting from the washing and transport of incoming raw sugar beets may also contain crop pests, pesticides, and pathogens (IFC 2007).

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For treating SBPW, a combination of different methods is used. Filtration, flow and load equalization, sedimentation, biological treatments (e.g., anaerobic followed by aerobic treatment and nutrient removal), chlorination, and composting are some of the common methods that are used (IFC 2007). Impoundments or lagoons are sometimes used for removing sediment-bound nutrients from wastewater as well as treating them before discharging (Calero *et al.* 2000; Fonade *et al.* 2000). However, they require a large surface area (Nahle 1998), could be a source of odor emission to the neighbor if not managed properly (Marden *et al.* 1980), and may cause groundwater contaminations (Nahle 1998; Wersin *et al.* 2001) and algal production due to excess amount of nutrients contained in wastewater (Nahle 1998). Biological treatments (e.g., combined anaerobic and aerobic treatments) are commonly used due to their high COD removal efficiency, lower sludge production, less space requirement, low odor emission, and energy recovery via anaerobic digestion (Nahle 1998; Austermann-Haun *et al.* 1999). Energy recovery from anaerobic processes, however, is not generally efficient, and the aerobic processes are highly energy intensive and produce a lot of solids which are costly to treat and dispose of (Wei *et al.* 2003; Rabaey & Verstraete 2005; Murray *et al.* 2008; McCarty *et al.* 2011). Therefore, researchers and scientists are in search of alternative cost-effective and less energy intensive technologies for wastewater treatment.

Currently, microbial fuel cells (MFCs) have shown promise for wastewater treatment, a technology that can synergistically mitigate organic and inorganic matters by converting them into electrical energy when treated under an anaerobic environment (Logan *et al.* 2006; Lovley 2006; Rittmann 2006). A number of studies have been conducted with different wastewaters as substrates for the MFC, such as domestic wastewater (Ahn & Logan 2010), dairy wastewater (Kelly & He 2014), starch processing wastewater (Kim *et al.* 2004), chocolate industry wastewater (Patil *et al.* 2009), potato processing wastewater and olive mill wastewater (Durruty *et al.* 2012), brewery wastewater (Feng *et al.* 2008), sanitary landfill leachate (You *et al.* 2006), sewage sludge (Yuan *et al.* 2012), cane sugar mill wastewater (Kumar *et al.* 2016), and swine wastewater (Min *et al.* 2005), which have shown the potential for wastewater treatment and electricity generation.

Performance of the MFC for wastewater treatment and electricity generation depends on the type of substrate and its concentration, MFC's configuration, and treatment conditions. Generally, pure substrates generate higher power densities than actual wastewater with complex organic substrates. Liu

& Logan (2004) obtained power densities of 494 mW m^{-2} using glucose compared to 146 mW m^{-2} using domestic wastewater in a single-chamber air-cathode MFC. Feng *et al.* (2008) obtained an increased power density of 205 mW m^{-2} from brewery wastewater, increasing the strength of the wastewater. In contrast, the highest volumetric power density of 11.6 W m^{-3} was observed at $4.3 \text{ kg COD m}^{-3} \text{ d}^{-1}$ loading rate, but the COD removal from leachate was the highest, 89.4%, at $0.65 \text{ kg COD m}^{-3} \text{ d}^{-1}$ (Zhang *et al.* 2008). These studies demonstrated that the power generation and treatment effectiveness of any substrate cannot be predicted *a priori*.

There is limited research where sugar beet derived wastewater was used as a substrate for MFC. Sikora *et al.* (2011) used sugar beet molasses and ferric oxide to select bacterial consortia capable of dissimilatory Fe(III) reduction. Güven *et al.* (2009) used an electrochemical process to treat simulated sugar beet wastewater and observed a significant reduction of COD. However, no MFC research is reported with SBPW that focused on both treatment (e.g., pollutants removal rate, optimum loading rate, and types of microorganisms involved) and electricity generation. Although there are many types of research with MFC for wastewater treatment, the optimum MFC configuration is still uncertain for actual field applications. Therefore, this study aimed to develop an inexpensive MFC system (without using chemical mediator or catalysts) to generate electricity and to treat SBPW at the same time. Optimum treatment conditions for solids and organic material removed from the SBPW and morphology of microorganisms involved were also investigated.

MATERIALS AND METHODS

Wastewater characteristics

The SBPW was collected from the wastewater treatment plant of American Crystal Sugar Company, located in Moorhead, MN, USA ($46^{\circ}54'05.1''\text{N}$, $96^{\circ}45'34.5''\text{W}$). The wastewater was collected from the primary clarifier overflow, which had the COD concentration of $14,895 \text{ mg L}^{-1}$ and a pH of 6.1. Other characteristics of the wastewater are presented in Table 1. Ammonia nitrogen ($\text{NH}_3\text{-N}$) concentration was very low in the SBPW (Table 1) as an inhibitory substance, indicating the suitability of this wastewater for producing electrical power (Feng *et al.* 2008). The

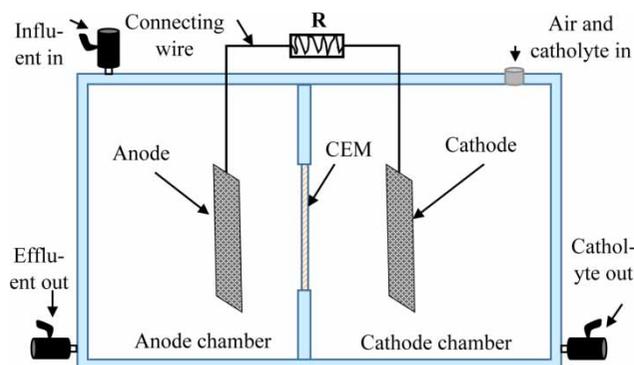
Table 1 | Raw SBPW wastewater characteristics

Parameter	Unit	Value
pH	–	6.1 ± 0.03
COD	mg L ⁻¹	14,895 ± 738
Total organic carbon	mg L ⁻¹	5,800 ± 707
Total suspended solids	mg L ⁻¹	1,140 ± 158
NH ₃ -N	mg L ⁻¹	1.42
Total hardness	mg L ⁻¹	5,545
Total volatile suspended solids	mg L ⁻¹	100

collected wastewater was stored in a refrigerator at 4 °C until use.

MFC configuration and operation

The dual-chamber MFC (Figure 1) used in this experiment was constructed with clear PVC plastic. Although the single-chamber MFC is generally more efficient in power production, the dual-chamber MFC is often preferred to investigate new substrates or techniques or reactions (Logan *et al.* 2006). The total volume of each chamber was 343 mL in which 280 mL was used as the working volume. The chambers were separated by a CMI-7000S cation exchange membrane (CEM) (Membrane International Inc., Ringwood, NJ, USA) to allow the flow of cations from anode to cathode chambers for completing an electrical circuit. The area of the CEM was 25 cm², and the membrane was used without any pretreatment except soaking in deionized water. Untreated non-wet and wet-proof (30%) carbon papers (MGL370, Fuel Cell Earth LLC., Stoneham, MA, USA) were used as the anode and the cathode, respectively. Wet-proof carbon paper was used because oxygen was used as the electron acceptor

**Figure 1** | Schematic diagram of a dual-chamber MFC (dimensions are not to scale).

(HaoYu *et al.* 2007). Both the anode and the cathode had equal surface areas of 1,250 mm² and they were installed 10 mm apart (i.e., 20 mm apart from each other) from the CEM and were held from the top by copper wires. The connection of the wires with carbon papers was covered by an inert insulating epoxy with conductive properties. The electrodes were connected to an external load by a resistance substitution box (Elenco model RS-400, Elenco Electronics, Inc., Wheeling, IL, USA).

In this study, total acclimation period was about 3 months and during the acclimation time, three batches of SBPW and anaerobic sludge were replaced for obtaining stable power output. All of the experiment was carried out with a COD concentration of 1,000 mg L⁻¹. After start-up, the MFC experiments were conducted at fed-batch mode under laboratory condition in room temperatures of 28 ± 2 °C. To investigate the effect of SBPW strength and to determine the optimum COD concentration for effective wastewater treatment and electricity generation, COD concentrations of the SBPW were varied (505, 1,000, 2,565, 4,783, and 5,750 mg L⁻¹) by diluting with deionized water. Anaerobic sludge collected from a thickening tank of the SBPW treatment plant was used to inoculate the anode chamber. Each time after the substrate replacement for a new batch of experiments, the headspace of the anode chamber was purged with nitrogen to expel oxygen. The cathode compartment was filled with 50 mM monobasic potassium phosphate (KH₂PO₄) salt solution as catholyte and was continuously purged with air using an airstone. However, no mechanical mixing was employed for any chamber except manual shaking occasionally. The pH of the catholyte was adjusted to 7.2 ± 0.1 by using 1 N NaOH solution.

Analysis and calculations

Voltage generation was measured and recorded continuously every 10 minutes using a data acquisition system (CR10x, Campbell Scientific, Logan, UT, USA) across a 1,000 Ω resistor. For constructing polarization curves, the load between two electrodes was varied in the range of 22 Ω to 47 kΩ for each substrate concentration. For each change of resistance, the resulting voltage between two electrodes was measured after the voltage had been stabilized (usually between 5 to 10 minutes). The current was calculated according to Ohm's law as follows:

$$I = \frac{U}{R} \quad (1)$$

where U is the voltage output measured by a data logger (mV), I is the current (mA), R is the external resistance (Ω). The calculated current was divided by the projected surface area of the electrode to estimate the current density. From the calculated I (Equation (1)) and measured U , the power density, P , was calculated as follows:

$$P = \frac{IU}{A} \quad (2)$$

Subsequently, from the polarization curve, maximum power density, P_{\max} , and internal resistance were obtained. According to Ohm's law, internal and external resistances are the same at maximum power density. The Monod equation was used to estimate the maximum cell voltage and half-saturation constant by using CurveExpert Basic Software (Version 1.4, Daniel G. Hyams). Coulombic efficiency (CE) was calculated as follows:

$$CE = \frac{CP}{CT} \times 100 \quad (3)$$

where CP = total coulombs calculated by integrating the current over time as

$$CP = \int I dt \quad (4)$$

CT = theoretical amount of coulombs that is available from COD, which was calculated as

$$CT = \frac{FbV\Delta COD}{M} \quad (5)$$

where

F = Faraday's constant ($96,485 \text{ C mol}^{-1}$ of electrons),

b = number of moles of electrons produced per mole of substrate ($b = 4$),

V = liquid volume (mL),

ΔCOD = COD concentration difference (g L^{-1}), and

M = molecular weight of the substrate ($M = 32$).

The MFC experiment with a particular substrate concentration was continued until the voltage generation dropped around 10 mV. Thereafter, the anodic liquid was replaced for a new batch with different substrate concentration. Influent (before MFC-treatment) and effluent (after MFC-treatment) samples from the each batch of the experiment

were collected and analyzed for COD, total suspended solids (TSS), pH, and electrical conductivity. COD was measured by a DR 2800 spectrophotometer and a COD 0-1,500 mg L^{-1} range kit (Method 8000; Hach Company, Loveland, CO, USA). TSS, electrical conductivity, total organic carbon, $\text{NH}_3\text{-N}$, total hardness, total volatile suspended solids, and pH were measured according to *Standard Methods* (APHA 2005). All analyses were done in duplicate, except a few instances in triplicate. COD and TSS removal efficiencies were calculated as follows:

$$\eta_{COD} = \frac{COD_{inf} - COD_{eff}}{COD_{inf}} \times 100 \quad (6)$$

$$\eta_{TSS} = \frac{TSS_{inf} - TSS_{eff}}{TSS_{inf}} \times 100 \quad (7)$$

where

η_{COD} = COD removal efficiency in percent,

η_{TSS} = TSS removal efficiency in percent,

COD_{inf} = influent COD concentration,

COD_{eff} = effluent COD concentration,

TSS_{inf} = influent TSS concentration, and

TSS_{eff} = effluent TSS concentration.

Scanning electron microscopy

Scanning electron microscopy (SEM) analysis was carried out to investigate the morphology of the biofilms grown on the anode surface of the MFC. At the end of the experiment, a few pieces of anode samples were cut by a sterile scissor and further processed for acquiring SEM images. Electrodes were fixed in 2.5% glutaraldehyde (Tousimis, Rockville, MD, USA) in sodium phosphate buffer, rinsed in buffer and deionized water, and dehydrated with a graded alcohol series (15 minutes each in 30%, 50%, 70%, 90%, and two changes of 100% ethanol). They were critical-point dried in an Autosamdri-810 critical-point drier (Tousimis, Rockville, MD, USA) with liquid carbon dioxide as the transitional fluid. The electrodes were attached to aluminum mounts with adhesive carbon tape and coated with gold/palladium using a Balzers SCD 030 sputter coater (Balzers Union Ltd, Liechtenstein). Images were obtained with a JEOL JSM-7600F scanning electron microscope (JEOL USA, Inc., Peabody, MA, USA) operated at 15 kV.

RESULTS AND DISCUSSION

Polarization curves and maximum power density

Figure 2 demonstrates the generation of power from the two-chamber MFC that used SBPW at different COD concentrations. The highest cell voltage, 392 mV, was obtained for 1,000 mg L⁻¹ COD while the lowest, 223 mV, was obtained with 4,783 mg L⁻¹ COD (Figure 2(a)). The cell voltage gradually decreased with increased current density; however, at around 30 mA m⁻², the cell voltage dropped suddenly by a large amount (cell voltage drop varied from 55% to 63% from the maximum for different substrate concentrations). A similar trend was also observed for the power curve (Figure 2(b)). This phenomenon of sudden large voltage and power drops is known as power overshoot (Ieropoulos *et al.* 2010). The reason behind this type of power overshoot might have been the acclimation of the MFC to a low current density (higher external resistance) (Hong *et al.* 2011) and the electrical and ionic depletion effects (Ieropoulos *et al.* 2010). The MFC was acclimated to a low current density using a 1,000 Ω external resistor.

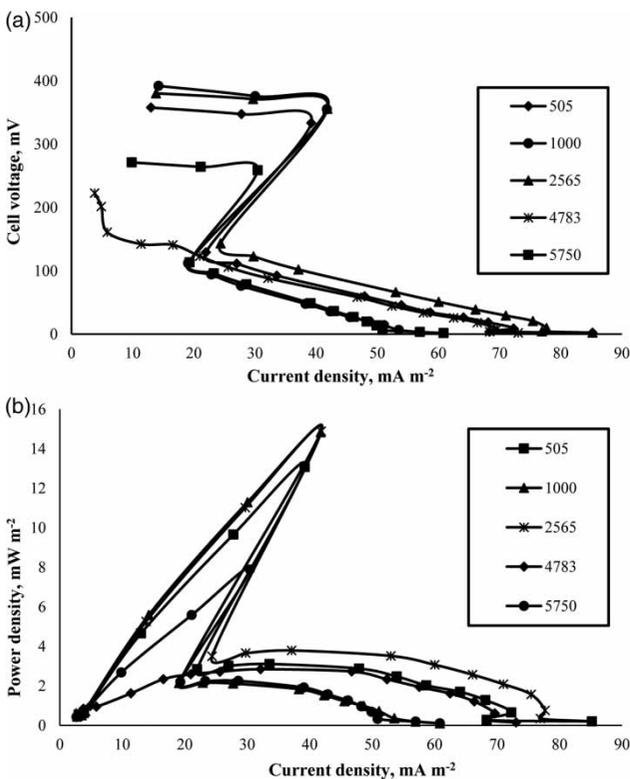


Figure 2 | (a) Polarization curve (cell voltage vs current density) and (b) power curve (power density vs current density) obtained in this study at different COD concentrations.

At the time of the polarization test, when the external resistor was shifted from the higher to the lower values (i.e., lower to higher current densities), the capability of the biofilm for a current generation was limited. At the lower resistor values, the demand for electrons (because of the increased conductive external path) exceeded the rate that the biofilm was able to supply. As a result, the anolyte was unable to supply electrons and ions in sufficient quantities to maintain the rate of power production (Ieropoulos *et al.* 2010; Hong *et al.* 2011). However, the microbial community was likely able to adjust to the dynamic and changing conditions in the anode chamber as it continued to generate power after the disappearance of overshoot, as shown in the Figure 2(b).

The maximum power density produced by the MFC varied with substrate concentration. The power density increased up to 2,565 mg L⁻¹ COD concentration and decreased thereafter. The maximum power output (determined from the power curve) measured was 14.9 mW m⁻² at 2,565 mg L⁻¹ COD concentration. This power density is much lower than that reported by other researchers. As mentioned previously, Liu & Logan (2004) obtained power densities of 494 mW m⁻² using glucose compared to 146 mW m⁻² using domestic wastewater, in a single-chamber air-cathode MFC. Feng *et al.* (2008) obtained an increased power density of 205 mW m⁻² from brewery wastewater, increasing the strength of wastewater. The differences in power density in this study and others are likely due to the differences in substrate, MFC configuration, and the presence of power overshoot which might have caused underperformance in terms of generating maximum power output (Nien *et al.* 2011).

As previously mentioned, maximum power densities were found to increase with increasing substrate concentration up to 2,565 mg L⁻¹. This was probably due to the sufficient carbon sources and increased conductivities as suggested by Zhang *et al.* (2009). However, the maximum power production decreased as substrate concentration increased to 4,783 and 5,750 mg L⁻¹ COD, indicating that an optimization of the substrate COD concentration and bacterial density or population is critical for the maximum power output (Masih *et al.* 2012). Our finding from this study is also consistent with other past studies in which the maximum power production decreased after a certain substrate loading rate (Wang *et al.* 2006; Nam *et al.* 2010; Masih *et al.* 2012; Wang *et al.* 2016). The presence of colloidal particles in real wastewater could have worked as rate-limiting and resistance-increasing factors (Wang *et al.* 2016), which might have a negative effect on power

production with increasing substrate concentration after a certain value. Also probably at higher substrate concentration, oxidation ability of electrogenic microorganisms went beyond their capacity and the substrates seemed to be utilized by the other anaerobic microorganisms in the anode compartment (Behera & Ghangrekar 2009). Furthermore, long-term use of the MFC could introduce biofouling on the membrane, which might have limited the mobility of protons, increased internal resistance, and, as a result, decreased the MFC performance (Choi *et al.* 2011).

Despite underperformance of the MFC due to overshoot problem, the power production from the SBPW was comparable to many studies although a number of studies outperformed it (Table 2). Performance of the MFC depends on several factors, such as MFC-architecture, substrate types and concentration levels, bacteria types and population, materials used, and experimental conditions, which makes exact comparison among studies very difficult (Liu *et al.* 2005; Feng *et al.* 2008). The majority of the studies included in Table 2 used a performance enhancer (e.g., use of Pt catalyst for electron transfer in single-chamber architecture) and produced power outputs in the order of 1,410.2 mW m⁻² (Zhang *et al.* 2009) and 334 mW m⁻² (Ahn & Logan 2010), which are much greater than the present study. In contrast, without any modification of real wastewater except dilution or using performance enhancer or architecture, this study produced power densities higher than that obtained by Du & Li (2017) and Kargi & Eker (2007). This holds promise for more power production from the SBPW if catalysts were used for electron transfer and for reducing potential oxygen diffusion.

Coulombic efficiency

CE was observed to be low and ranged from 0.73% to 6.21% (Figure 3). A low CE of 8% was reported when starch processing wastewater was used in an air-cathode MFC constructed with a CEM (Lu *et al.* 2009). The highest CE of 6.21% was observed at 505 mg L⁻¹ COD concentration and decreased with increasing COD concentrations. Often, for real wastewater with complex substrates, CE is found to be widely varied (Table 2). A number of factors were believed to be responsible for the lower CE. Although they were not measured in this study, a part of the substrates in the anode chamber was degraded by the oxygen which was diffusing through the CEM (Nam *et al.* 2010). The amount of oxygen diffused was likely to increase with the increased treatment time taken for degrading more

Table 2 | Comparisons of performances between the MFC in this study and other similar studies using substrates of different origins

Substrate type	MFC type	Membrane materials	Catalyst used	Initial COD, mg L ⁻¹	COD removal, %	Current density, mA m ⁻²	Maximum power density, mW m ⁻²	Coulombic efficiency, %	Reference
Beet sugar factory wastewater	Dual	CEM	None	500–5,750	73–97	61–85	2.84–14.9	0.73–6.21	This study
Starch processing wastewater	Single	PEM	Pt	4,852	98	893	239	8	Lu <i>et al.</i> (2009)
Waste activated sludge and solid potato	Dual	CEM	PBS	1,569–4,245	39.5–89.6	5–150	0–6.80	0.30–43.6	Du & Li (2017)
Cane sugar mill effluent	Dual	PEM	KmNO ₄	3,605	56	50	140	72	Kumar <i>et al.</i> (2016)
Molasses wastewater	Single	NA	NA	1,020–9,400	43–95	71–76	11.6–13.5	NA	Lee <i>et al.</i> (2016)
Molasses wastewater	Single	NA	Pt	NA	30	4,947.9	1,410.2	≈1	Zhang <i>et al.</i> (2009)
Domestic wastewater	Single	NA	Pt	800–900	88	977	334	47–57	Ahn & Logan (2010)
Swine wastewater	Dual	PEM	Pt	8,250	83	150	45	NA	Min <i>et al.</i> (2005)
Urban wastewater	Dual	Salt bridge	NA	NA	44	180	25	25	Rodrigo <i>et al.</i> (2007)
Fermented wastewater	Single	NA	Pt	1,920–4,800	93	NA	600	<1	Nam <i>et al.</i> (2010)
Synthetic molasses wastewater	Dual	Salt bridge	NA	1,000–6,000	80	50	2.9	NA	Kargi & Eker (2007)

CEM, cation exchange membrane; PEM, proton exchange membrane; PBS, phosphate buffer solution; na, not available.

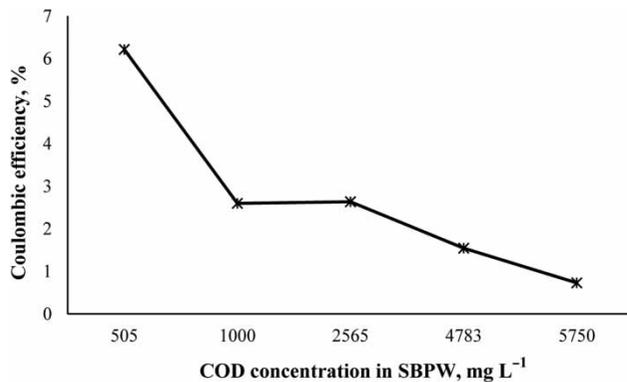


Figure 3 | CE of the MFC at different COD concentrations.

substrates, which resulted in lower CE (Min *et al.* 2005). Low coulomb recovery could also be due to the fermentation and methanogenesis consumptions of coulombs, the growth of biofilm, and the aerobic respiration of the biofilm by diffused oxygen (Zhang *et al.* 2009; Nam *et al.* 2010).

COD removal efficiency

The MFC system was found to be highly effective in removing COD from the SBPW (Figure 4). The highest COD removal was 97.1% at 4,783 mg L⁻¹ COD concentration and the lowest was 73.3% at 505 mg L⁻¹ COD concentration. In this study, COD removal rates increased with increasing COD concentrations as shown in Figure 4 and indicated no process inhibition due to increased COD concentration in the MFC system (Mohan *et al.* 2009). Feng *et al.* (2008) observed a similar COD reduction up to 98% when treating brewery wastewater in MFC, which is in good agreement with that attained in this study. Further review of the literature, as presented in Table 2, suggested

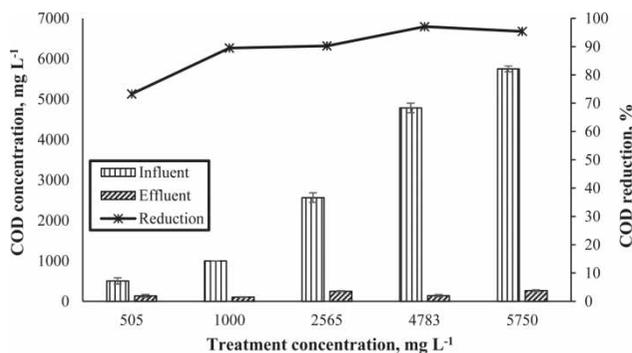


Figure 4 | COD removal efficiency of the MFC at different COD concentrations (the error bar represents standard deviation).

a wide range of COD removal using the MFC. Differences in reactor architecture, operational conditions, and substrate concentrations probably resulted in the variation of COD removal efficiencies across different studies (Kim *et al.* 2016).

Substrate removal and cell voltage generation

The maximum cell voltage generation as a function of initial substrate concentration (S) appeared to follow a saturation-type relationship (Monod kinetics) at lower substrate concentrations (Figure 5). The predicted maximum cell voltage, (V_{max}), was 294.8 mV with a half-saturation constant, K_s , of 43.5 mg L⁻¹ COD. The value of the half-saturation constant is lower than that obtained from glucose (79 mg L⁻¹; Liu & Logan 2004) and butyrate (93 mg L⁻¹; Liu *et al.* 2005). A lower half-saturation constant indicates that the system could produce more power in the lower substrate concentrations (Zuo *et al.* 2006). From Figure 5, it is also observed that the points on the graph for high substrate concentrations deviate remarkably from the model estimated line, suggesting that the substrates degraded at higher concentrations were not utilized for power generation as effectively as in the lower concentrations. This result is consistent with the study of Kim *et al.* (2016) in which they found that the rate constant for COD removal from the high strength wastewater was inconsistent with the one for low strength wastewater, likely due to the differences in removal mechanism.

TSS removal efficiency

The influent and effluent concentrations of TSS and its removal efficiency (with respect to the influent TSS

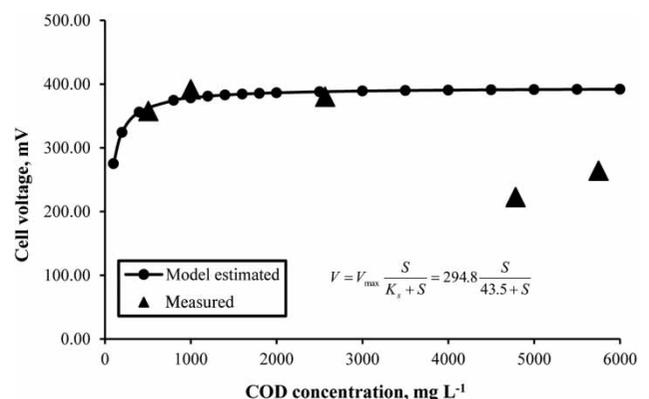


Figure 5 | Relationship between influent COD concentration and maximum voltage generation of the MFC operating on beet sugar factory wastewater.

concentration) are presented in Figure 6. High TSS removal efficiencies were observed in treating SBPW in the MFC and were ranged from 46 to 100%. The complete removal of TSS was observed for 505, 2,565, and 4,783 mg L⁻¹ COD concentrations, whereas the lowest (46.4%) TSS removal was

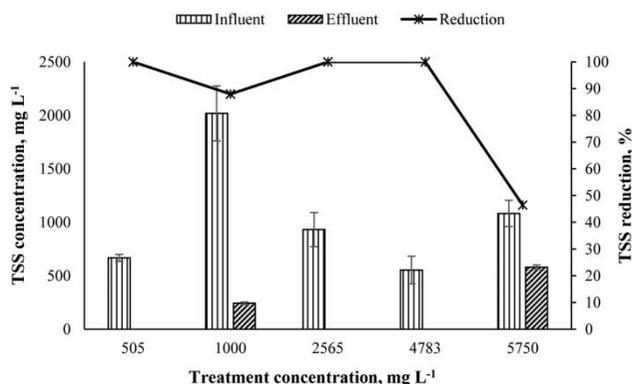


Figure 6 | TSS removal efficiencies of the MFC at different COD concentrations (the error bar represents standard deviation).

observed for 5,750 mg L⁻¹ COD concentration. This removal is higher than that observed for an upflow MFC that used seafood processing wastewater, where the TSS removal was in the range 45%–55% for different organic loading rates (Jayashree *et al.* 2016). High TSS removal could be attributed to the accumulation of fermentation products, including various volatile fatty acids (Jayashree *et al.* 2016) because the pH of the anodic liquid was observed to decline (data not shown). High TSS removal implies the production of less sludge, which is advantageous compared to aerobic treatment because the sludge handling process is high energy demanding.

Microbial community analysis

SEM micrographs were taken by cutting a small portion of the carbon paper anode used in the MFC and are shown in Figure 7. It is observed from the Figure 7(a) that a thick biofilm of densely packed bacterial cells was formed on the anode surface, covering the majority of the area. A

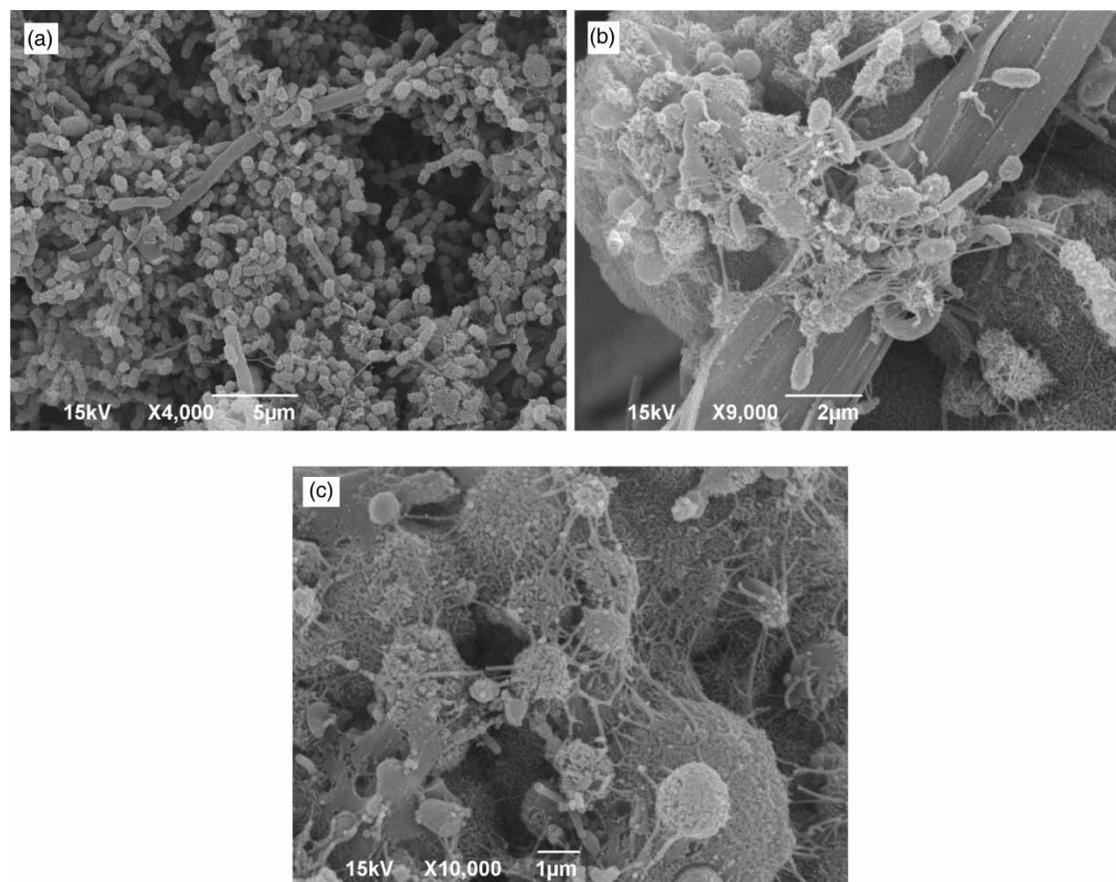


Figure 7 | SEM micrographs taken at (a) low ($\times 4,000$), (b) high ($\times 9,000$), and (c) high ($\times 10,000$) magnifications of carbon paper anode used in the MFC for SBPW treatment and electrical power generation.

diverse community of microorganisms was active for organic material degradation and electron transfer as seen from the Figure 7(b). Bacteria with different morphological structures and shapes, such as coccus, bacillus, and vibrio, were attached to the anode surface. Flagella, pili-like structures, and filamentous appendages were also observed in the magnified micrograph. An abundance of pili-based or nanowire-like bacterial structures indicates the potential mechanisms of electron transfer. Overall, the SEM micrographs suggested synergistic interactions among different microbial consortia for electricity generation as well as the treatment of SBPW for pollutants removal. However, further research is needed to identify the dominant genera for organic matter degradation and power generation.

CONCLUSIONS

The dual-chamber MFC was able to successfully generate electricity and simultaneously removed pollutants from the SBPW. The highest power density of 14.9 mW m^{-2} was attained at $2,565 \text{ mg L}^{-1}$ COD concentration despite the limitations imposed by the power overshoot. CE was low, indicating the diffusion of oxygen through the CEM from the cathode to the anode chambers and the occurrence of methanogenesis and fermentation processes. Over 97% removal of COD and up to 100% removal of TSS were achieved. A diverse community of microbial consortia was active for electricity generation and wastewater treatment. Further research is needed to increase power density and up-scaling the system for cost-effective power production and treatment.

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